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A NEW, LOW-ELEVATION APPALACHIAN SAXIFRAGE (*HYDATICA* NECKER EX. GRAY 1821) IN SOUTH CAROLINA

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A NEW, LOW-ELEVATION APPALACHIAN SAXIFRAGE
(*HYDATICA* NECKER EX. GRAY 1821)
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ABSTRACT

In 2002 a unique population of *Hydatica* Necker ex Gray 1821 (Appalachian Saxifrage) was discovered by Patrick McMillan in the upper Piedmont of Pickens County, SC, growing on a low-elevation granite dome within the Nature Conservancy's Nine Times nature preserve. The plants appeared to be similar to the widespread *Hydatica petiolaris* (Raf.) Small populations of the Blue Ridge Escarpment (BRE). However, the Pickens County population displays an annual life form and flowers during the late winter and early spring, in contrast to the perennial life forms and late spring through early fall flowerings of the BRE population. A study was initiated to determine if the population in Pickens County meets the criteria of the biological, phylogenetic and ecological species concepts and thus constitutes it as a novel species. Morphology comparisons, non-coding DNA analysis, common garden experiments, and ecological comparisons were chosen as species concept criteria to test high-elevation *H. petiolaris* populations and the low-elevation, Pickens County *Hydatica* sp. population. The floral form and stature, distinct DNA phylogeny, and unique adaptive zone of the Pickens County population proved distinct from other *Hydatica petiolaris* populations. Therefore, experimental evidence indicates that the Pickens County population meets three different species concepts, representing a novel species known only to this location.

DEDICATION

Thanks to my wife, Larisa Jean, and my children, Alekzandre, Lili, and Phillip, for their support and putting up with 3 years of fieldwork, and fridges full of plants, bugs, and sometimes dead animals; your love, patience, collaboration, and faith in my ability as a Husband, Father, and Student helped to drive me thus far.

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Thanks to my family, who often hiked with me searching for a small glimpses of this plant in the most rugged and beautiful of places, and to my hiking companion, Chad Bates, for making the long, lonely hikes into the granite domes of God's country more enjoyable. Mostly, I thank my Lord and Saviour, Christ the King, who has made all things known and unknown, and can still make Man anew. The hand that has drawn the limits of the granite hills and the boundary of the salted sea, still draws men to the foot of a Cross; Romans 10:9, The Holy Bible.

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CHAPTER ONE

INTRODUCTION

The Blue Ridge Escarpment.

The Blue Ridge Escarpment (BRE) is an east to southeast-facing escarpment that spans nearly 500 km between Georgia and Virginia, separating the Blue Ridge Valley and Ridge Highlands from the low-relief Piedmont and Coastal Plain (Spotila, et al. 2004) (Figure 1). It is composed of easily eroded and weatherable lithology that is rich in massive and resistant gneiss and metasedimentary rock and has relief exceeding 1,100 m (Hack 1982). Approximately 14% of Pickens County, South Carolina (SC), is composed of the BRE, with elevations ranging from 425 meters to 1,083 meters (m) above sea level (ASL), and average elevations at 580 m ASL (USDA 1972). These mountains feature diverse ecosystems composed of exposed to forested geologic features ranging in slope, aspect and elevations (Griffin 1973).

Within these mountainous areas are exposed “granite outcrops” surrounded by the various forest ecosystems. These outcrops support unique edaphic conditions, with rare and endemic vegetation not found in other regions (Wyatt and Fowler 1977, Murdy 1966). Some of these forest ecosystems are considered natural relics of former, early postglacial forests that occupied the BRE since the Holocene but now are limited to lower piedmont areas (Crosby and Anderson 1967).

Granite outcrops are composed mostly of granite and granite gneisses present and frequently occur as extensive areas of exposed rock (Burbanck and Platt 1964). The soils of these granite outcrops offer little nutrient support, due to the lack of a sustainable layer and existing shallow substrates that divide the outcrops into six specialized habitats: 1) bare rock; 2) rock crevices; 3) shallow depressions; 4) deep soil accumulations/vegetative mats; 5) seepage areas; and 6) wooded margins (Wyatt and Fowler 1977). They have high acidity and low calcium and phosphorus recruitment opportunities, and saprolite base layers with unique and historical soil-vegetative associations (Pittillo, et al. 1998).

Studies by Wyatt and Fowler (1977) note that characteristic species and associations can be found throughout the BRE on granite outcrops, with each habitat having its share of endemic species. Other studies have found that the shallow-soiled, well drained, sunny site conditions have led to speciation events in vegetation, producing populations endemic only to outcrops with little to no opportunity for recruitment or dispersal of genetic material (Rajakaruna 2004).

The bare rock is typically populated by crusteous lichens, such as *Cladonia* sp. and *Heterodermia* sp. (McCune, et al. 1997) that can tolerate the extreme heat and the lack of moisture at these locales. Rock crevices offer a more defined surface for root growth and soil accumulations, where seeds of neighboring forests lodge themselves and germinate. Species that seem to tolerate the extreme conditions include *Juniperus* sp. and *Pinus* sp. (Wiser, et al. 1996). The shallow depressions are often dominated by *Diamorpha smallii* Britton ex Small, a diminutive, annual, red-pigmented succulent. On deeper soils other annuals, such as *Minuartia glabra* (Michx.) Mattf., can be found (Spira 2011). Deeper soil accumulations develop vegetative mats, mostly colonized by mosses, such as *Grimmia laevigata* (Brid.) Brid.. These vegetative mats offer substrate for trees and a variety of annuals to establish seedlings and often herbaceous perennials. Seepage areas offer a unique composition of wetter species, often colonized first by mosses, such as *Sphagnum*, *Campylopus*, and *Polytrichum* spp., and then by various annuals and perennials. The woodland margins are typically composed of an oak-hickory complex of shallow soil types (10cm – 50cm).

The case of Hydatica petiolaris *(Raf.) Small*

The family Saxifragaceae contains mostly perennial herbs with a basal rosette of leaves, rarely cauline, with actinomorphic or zygomorphic flowers in a panicle or cyme (Radford, et al., 1968). The range of the Saxifragaceae are mostly throughout the greater part of the temperate and sub-arctic zones of the Northern Hemisphere. Many are found in the alpine zone or persisting as alpine relics within glacial refugia (Comes and Kadereit 1998). Many members of the Saxifragaceae are possibly artic disjuncts that survived in higher elevations since the last ice-age (Wiser 1994).

The evolutionary history of the *Saxifragaceae* represents one of the major phylogenetic difficulties within angiosperms (Soltis, et al. 1990). The family is known to have floral polymorphisms with no resolved correlations between polymorphisms and elevation or microclimate (Bensel and Palser 1975, Stevens 1988, Soltis and Soltis 1989) (Figure 3). Current studies show no clear relationship between elevation and petal arrangement (Folk 2013, Malo and Baonza 2002). Elevation plays a key role in both sexual reproduction phenology and physiological performance of individual populations, in that, higher elevation sister taxa tend to flower later in spring and are heterosexual, whereas low-elevation sister taxa tend to flower earlier and are self-fertilizing (Mazer, et al. 2010). Also, environmental factors have been shown to affect reproductive strategies of sister taxa that inhabit similar and close proximity ecotones (Macnair, et al. 1989).

A regional BRE saxifrage, *Hydatica petiolaris* (Raf.) Small, known as "Cliff Saxifrage," is found at high-elevations as a perennial, occupying moist rock outcroppings, crevices in exposed rock outcrops, periglacial boulderfields, and rocky seeps/spring seepage slopes (Radford, et al. 1968, Weakley 2012). It flowers from June to August, and is a Southern Appalachian endemic (Weakley 2012). Plants are usually robust, with flowers that have 5 petals in a zygomorphic arrangement (Figure 4, A). The 3 upper petals are distinctly clawed with 2 yellow spots. The 2 lower petals are smaller, cuneate, and not spotted. It has a basal rosette of simple leaves, with coarsely dentate leaf margins and is typically found above 426 meters (m) above sea level (asl) (Radford 1968, Weakley 2015).

In 2002 a unique population of *Hydatica* Necker ex Gray 1821 (Appalachian Saxifrage) was discovered by Patrick McMillan growing in the upper Piedmont of Pickens County, SC, growing on a low-elevation (<426 msl) granite dome. The plants appeared to very similar to the widespread *Hydatica petiolaris* but display an annual life form and flower during the late winter and early spring. The low-elevation population is diminutive with similar rosette of simple, coarsely dentate leaf margins, but it presents a nearly actinomorphic arrangement of 5 clawed petals, with all petals bearing 2 yellow spots (Figure 4, B).

Species concepts

The question as to what distinguishes one species from another has been one throughout the history of science. The earliest attempt to define a species was by John Ray in his 1686 publication, *History of Plants*, where he stated that “one species never springs form the seed of another nor vice versa” (Mayr, 1982). Following thereafter, Carl von Linnaeus (1753) formalized taxonomy within his work, *Species Plantarum*, by cataloging species into a two-part naming system, based on morphology. From there, the debate long ensued to clarify what truly is meant by species, resulting in Darwin’s *Origin of the Species* (1859) proposing “stages” in speciation.

Then in the early 1920s and 1930s Theodosius Dobzhansky and Ernst Mayr, began to articulate the genetic processes that occur when incipient species begin to diverge, with Dobzhansky describing the critical role for the formation of new species, reproductive isolation (Mallet, 2004). Ernst Mayr’s ideas had importancefor the development and acceptance of the modern metapopulation lineage concept of species (De Queiroz, 2005).

From this early history of thinking, three major contemporary concepts arose to categorize species: the Biological Species Concept (BSC), Ecological Species Concept (ESC), and Phylogenetic Species Concept (PSC). These three major concepts can conform to a single general concept under which species are equated with separately evolving metapopulation lineages;, however for study of individual population speciation, the three are segregated (De Queiroz, 2007). All of these concepts have strengths and flaws; therefore there is no consensus about which species concepts should be used, particularly in plants (Judd et al. 2008).

Systematics

Morphological differences are a central focus for naturalists, paleontologists, and museum taxonomists, whereas genetic differences are key for population geneticists and molecular systematics (De Quirez, 2007). Systematics embodies these differences and provides a framework to study the historical aspects of evolution (Schuh and Brower, 2009). The diversities aid in segregating individuals into *taxa*, which possess the property of monophyly, defined as all of the descendants of a most recent common ancestor, which share the same traits or characters (Judd, et al., 2008; Simpson, 2006). It builds on three principles: 1) the recognition of basic units of biological diversity in nature, 2) classification of those units into a hierarchical scheme that reflects their phylogenetic relationships, and 3) the placement of the information about species and their classification in a broader context that helps to define their relations to one another and to a broader scale (Schuh and Brower, 2009). From this framework, individual populations of taxa can be studied and placed within a phylogenetic framework that best gives the most likely pattern of descendance among its members, through observable differences (Skelton, 2002).

Objectives of Study

This research focuses on three objectives: (1) to test the BSC, ESC, PSC of the low-elevation population and determine if it constitutes a novel, undescribed species; (2) to look for molecular differences between the low-elevation and high-elevation populations; and (3) to examine the evolutionary relationship of the low-elevation population to other known

Hydatica species within the BRE. The first hypothesis proposes that the low-elevation *H. petiolaris* population, hereafter referred to *Hydatica* sp.1, is actually a distinct taxon from the regional high-elevation populations, based on differences in morphological, ecological, and molecular characters. The second hypothesis proposes that the molecular variation is significant among genetic regions of the individual populations to constitute a distinct taxon within the BRE.

CHAPTER TWO

MATERIALS AND METHODS

The Nine Times Nature Preserve (Study Site).

The Nine Times Preserve (NTP), owned and managed by The Nature Conservancy (TNC), holds a unique granite outcrop within the Pickens County portion of the BRE. The NTP is a 560 acre tract that holds five mountains, four forest types, and a designation by TNC as a "southeastern wildflower showcase," due to its support of unique and endemic floral populations (The Nature Conservancy 2012). It ranges in elevation from 365 m to 520 m ASL with granite outcrops composing 3.97 hectares (1.75%) of the total area at NTP (Figure 2).

The vegetation at NTP granite outcrop is composed of a typical mosaic of outcrop vegetation, with the bordering forests and woodland associations composed of an oak-hickory complex interspersed with conifers and junipers. The aspect of the study site is south-facing with the majority of slopes varying from gently sloping (1:6 ratio) to steep terrain (1:1 ratio). Temperatures at the site show summer highs exceeding 44º C and winter lows at -4º C. Hydrology on the site is mainly supported by spring seeps in the vegetated areas and by seasonal rainfall throughout the year.

Study sites and sampling

Voucher specimens from the Clemson University Herbarium (CLEMS) were examined to compare floral morphologies of *H. petiolaris* with photographs and whole plant vouchers taken at Nine Times Preserve, SC (NTP). High-elevation study sites were chosen from vouchers that were of equidistance apart, ranging from 30 to 40 kilometers of geographic distance that bore populations of the same morphological characteristics and elevation for comparison to the low-elevation study population (Table 1). Specimens, seeds, and tissue samples from *Hydatica* spp. were collected from five study sites: Nine Times Preserve, SC (NTP), Blood Mountain, GA Region (BM), Tallulah Gorge, GA Region (TG), Big Creek Trail, GA (BCT), and the Balsam Mountains region, NC (BMT) (Figure 5). The material was used for new vouchers, morphological analysis, common garden plantings, and genetic analysis. The BMT and NTP sites were used for microclimate data, ecological analysis, and phylogenetic reference. This strategy yielded a matched dataset of localities from which I had morphological, physiological, genetic, macroclimate, and microclimate data.

Biological Analysis

Four 1-meter square study plots were established at the NTP with a 1 cm grid applied to each, and 10 germinating individuals were selected, via a random number generator in MS Excel (Figure 6). Plots 1 and 3 included individuals that persisted from the previous year's growing season. The selected individuals within each plot were monitored for leaf and flower traits, flowering times, and fruiting times between February and May of 2013 and 2014. Soil depth at each plot was also recorded for each individual. The BM, TG, BCT, and BMT sites were visited during recognized flowering and fruiting times to verify species presence and to collect morphological and floral characteristics (Radford, et al. 1968; Weakley 2012).

During flowering times, 30 flowers each, from the NTP and BMT locations, were live collected and petals were measured using a modified methodology as described by Whitney et al. (2012), here explained. Petals were removed from flowers, using dissecting tweezers and a Leica Stereo Microscope, gently stuck to a glass microscope slide labeled according to location, and covered with transparent sticky tape (Figure 7). The petals were mounted as flat as possible. Each slide was then read on a Leica DM500 microscope, after calibrating the eyepiece graticule with a stage micrometer (Leica, 2004). Measurements were taken of the petal length and the petal width to the nearest 0.1 mm. Mensural data was stored in Microsoft Excel 2010 and later exported into csv format for use in R statistical software.

Controlled Garden Experiments

A design was conducted at a both low and high-elevation gardens to determine if location bears an influence on flowering time: the SCBG, and a private residence in Cashiers, NC. Seeds collected from the study sites were sown in 10-cm square plastic pots filled with peat, divided into two hydrologic groups (ambient and scheduled watering). They were exposed to outdoor temperatures at both sites and monitored to assess potential biological isolation via differential phenology, should the two populations exist in sympatry. Photographic documentation recorded germination and flowering times for each sample set during the 2013-2014 growing season (Figure 8).

Ecological Analysis

The regional macroclimate and local microclimate regimes of study sites was documented with macroclimate data using ArcGIS 9.3, while microclimate data at the BMT and NTP study plots was recorded with “HOBO monitors” (Onset Computer Corp, Pocasset, Mass). Monthly temperature, barometric pressure and windspeed with direction from the National Weather Service’s National Digital Forecast Database (NDFD) for 2 localities (BMT and NTP). These variables summarize temperature and precipitation data from weather stations from 1950 to 2000, spatially interpolated to localities between weather stations, at a resolution of approximately 1 km^2. The microclimate data were retrieved from the “HOBO” monitors (Onset Computer Corporation), which recorded temperature and relative humidity at half-hour intervals. The data were collected continuously over the duration of the study (> 1 year).

The natural community structure of the study sites was documented using a modified Carolina Vegetation Survey plot system (Peet et al. 1998). Two, 30-m^2 survey plots were established at the high-elevation BMT site and ten 30-m^2 survey plots were established at the NTP. All plots were surveyed as intensive modules (Figure 9). Each intensive module was sampled individually for presence of vascular plant species in a series of ten, 1-meter parallel transects. Woody individuals taller than breast height (1.4 m) were tallied within each intensive module according to diameter (cm) at breast height (DBH).

Soil samples were collected from each of the two BMT high-elevation sites, from three NTP low-elevation sites. One sample from the South Carolina Botanical Gardens

(SCBG) served as a control, for a total of six samples. The soil samples from NTP and BMT sites were of minimal depth due to the granite substrate, with at least 380 cm^3 (8x8x5cm) collected for analysis. The samples were assayed for nutrients and texture by Clemson University Agricultural Service Laboratory, Clemson, SC.

Relationships between the environmental variables and the vegetative composition of individual sites were determined using R statistical software and the BiodiversityRGUI and Vegan packages (R Core Team 2014, Kindt and Coe 2005, Oksanen, et al. 2015). Ten NTP and two BMT sites were chosen for the CVS analysis, based on the elevations of these populations and amount of data available for each. Statistical inference about differences in the average number of species per site and average abundance per site was obtained from rank abundance curves and generalized linear model functions (R Core Team 2014). Rank abundance curves for the NTP and BMT sites were determined by calculating the total number of individuals for each species. Then the species were ranked from the most abundant to the least abundant. Finally a plot was constructed with the rank number on the horizontal axis, and the abundance on the vertical axis. For the GLM, a model was fitted with either *Hydatica* sp.1 or *Hydatica petiolaris* as the dependent variable, with soil depths, elevation, soil pH, P, K, Ca, Mg, Zn, Mn, Cu, B, and N as categorical explanatory variables, and a log link function and a Poisson variance function was applied, thus using model specifications that are realistic for count data (Kindt and Coe 2005). Then a Principal Components Analysis (PCA) model and Canonical Correspondence Analysis (CCA) model were employed to determine the relative importance and influences of the independent variables (environmental) on dependent variables (community and/or species composition), using a simple dataset of 3 NTP sites and 2 BMT sites, due to redundancies in data and lack of soils data for 7 NTP sites.

Phylogenetic Analyses

For comparative analysis of the study populations with family-level genera, effort was made to include as many taxa of each within the Saxifragaceae that best represented the family. A total of 101 sequences of various genera were obtained from GenBank. Not all genera within the family had sequences available; therefore, those with the most commonly shared loci but within the genus were obtained for the analysis. Regions of nDNA that were included in GenBank queries and acquisitions were Internal Transcribed Spacer (ITS) 1, 5.8s, and ITS2. These regions are the most commonly used in plant phylogenetics to infer evolutionary relationships within the Saxifragaceae (Soltis and Soltis, 1997) and within the various genera and sections (Soltis et al., 1996; Conti et al., 1999; Vargas, 2000). The acquisitions represent both local Blue Ridge genera as well as regional and worldwide genera.

Leaf material from individual study populations was collected from the highest elevation site, BMT, and from the lowest elevation site, NTP, then ground in liquid nitrogen with mortar and pestle, and extracted using Dneasy Plant mini Kit (Quigen). The ITS1-5.8s-ITS2 region was amplified by PCR using external primer pairs, via 5 ul of 10 FM N- ncl8S10 (forward primer, 5'-AGGAGAAGTCGTAACAA, anchored in 18S rDNA; Kuzoff, et al., 1999); 5 ul of 10 M C26A (reverse primer, 5'-GTTTCTTTTCCACCGCT, anchored in 26S rDNA) (Kuzoff, et al., 1999).

Fluorescent dye terminator sequencing reactions were performed using Illustra AutoSeq G-50 and same primer pairs, then viewed by electrophoresis, and sequenced on 3130xl genetic analyzer (Applied Biosystems Corporation), at both Western Carolina University (WCU) and the Clemson University Genomic Institute (CUGI). The primary reference for analysis was an evidence approach wherein the loci were compared to other sequences obtained from basic local alignment search tool (BLAST) on the National Center for Biotechnology Information (NCBI), U.S. National Library of Medicine's website (Altschul, et al. 1990). Sequence alignment was performed on the loci individually using Multiple Sequence Comparison by Log-Expectation in MUSCLE (Edgar, 2004) as implemented in MEGA version 6 (Tamura et al. 2013). The alignment was well accepted with few gaps in regions of the sequence. Despite the success of the alignment, it was post processed through the program GBLOCKS, which eliminates poorly aligned positions and divergent regions of an alignment of DNA or protein sequences (Castresana 2000). The use of GBLOCKS reduced the necessity of manually editing multiple alignments of the large datasets.

A best-fit evolutionary model was calculated in jModelTest (Posada 2008). With jModelTest, statistical selection of best-fit models of nucleotide substitution are possible. It implements five different model selection strategies: hierarchical and dynamical likelihood ratio tests (hLRT and dLRT), Akaike and Bayesian information criteria (AIC and BIC), and a decision theory method (DT) (Posada 2003). It also provides estimates of model selection uncertainty, parameter importances, and model-averaged parameter estimates, including model-averaged tree topologies.

Cladograms were constructed in MEGA version 6.0 using both Parsimonious Likelihood (PL) and Maximum Likelihood (ML) to estimate trees based on the results of the jModelTest computation (Tamura, et al. 2013), with Bayesian Inference (BI) and a coalescent species tree estimation approach implemented in the program BEAST2 (Bouckaert, et al. 2014), with clade support assessed by posterior clade probabilities.

The BEAST2 Markov chain Monte Carlo (MCMC) was run two times independently (each chain of 20 million generations) and combined for a total of 40 million generations to ensure convergence and high sample size, which were diagnosed in the program Tracer, version 1.6. Convergence was reached at 1 million generations, so the first 1 million generations were discarded from each chain as burn in. Posterior clade probabilities were plotted on the most likely tree. Most effective sample sizes of the posterior distribution (ESSs) were above 200, and some were above 1000. GTR-gamma was used to maintain comparability. A support cutoff of 50 was used for all bootstrap values, with posterior probabilities cutoff at 0.90.

CHAPTER THREE

RESULTS

Controlled Garden Experiments

The open garden experiments revealed flowering times were concurrent with each respective study plot despite the common garden environment (Table 2). Each garden plot bloomed corresponding to its study plot's population. Flowering times for Open Garden NTP plots occurred during mid-February in 2013 and 2014, while the Open Garden BMT populations began blooming in early May 2013 and 2014.

Climatic Data

Weather data from the World-Clim database and the National Weather Service's National Digital Forecast Database (NDFD) for Pickens, SC, and Balsam Grove, NC, were tested against the Hobo-Weather monitor data and found to have variations in the average monthly low temperatures and monthly average temperature. High temperatures did not vary between the two data sets. A comparison of the micro-climate weather data between the NTP and BMT sites found statistically significant variations between the two sites for monthly recorded data, with p-value < 0.0001(Table 3). The range of all data tested was January 13, 2014 to January 16, 2015 (Figures 10, 11, and 12). The average annual high and low at the NTP site were 29.59º Celcius (C) and 2.08 º C, respectively, while at the BMT site, the average annual high and low were 31.1 ºC and -3.27 ºC, respectively.

Carolina Vegetative Survey

Rank abundance curves are the easiest method of analyzing patterns of diversity (Kindt and Coe 2005). The abundance curves established for the CVS data for both the NTP and BMT sites, revealed diverse compositions of flora.

The NTP sites had associations composed mostly of rock, *Pinus virginiana*, *Juniperus virginiana*, miscellaneous mosses, and miscellaneous lichens (Figures 13 and 14, Table 4). The soils data determined that the vegetative mats within the study plots had an average soil depth of 26 mm. The soil chemistry data indicated the habitats were relatively “normal” in general chemistry with Mg being the predominant chemical and Ca following. The calculations found the base saturation of the soils to hold Ca at 12%, Mg at 13%, K at 1%, Na at 1%, for a total of 27% total base saturation

The BMT sites shared a similar abundance of rock, but *Sphagnum* sp., *Cyperus* sp., *Cladonia caroliniana*, *Grimmia* sp., and *Rubus* sp. were dominant in composition (Table 4). The soils data determined that the small amount of areas with accumulated soils had an average soil depth of 12 mm. The soil chemistry data indicated the habitats were also relatively normal in general chemistry with Mg being the predominant chemical and Ca following. The calculations showed the base saturation of the soils to hold Ca at 19%, Mg at 7%, K at 3%, and Na at 1%, for a total of 30% total base saturation. Potassium was excessive in these samples.

The CCA and PCA ordinations on the vegetative surveys revealed the dominant gradients controlling vegetation at multiple scales and identified frequency of species presence at each site.

The CCA revealed that soil depth had a strong influence on the presence of species, and that elevation is the second strongest factor explaining distributions among both high and low-elevation sites (Figure 15). The strongest gradients between the NTP and BMT sites was elevation, P, soil depth, and soil pH. However, the NTP sites often received equal or similarly weighted values among environmental variables. The CCA was applied to a reduced site matrix with only 3 NTP sites and 2 BMT sites being used for a more robust correlation. No soils data were available for 7 of the NTP sites, and thus those sites were excluded. This allowed the statistical software to identify which environmental factors were useless constraints. They were completely removed from the estimation, and no biplot scores or centroids were calculated for these constraints (R Core Team 2015).

An ANOVA of the explanatory variables showed p-values < 0.0001 for the elevation and soil depth, and for the P and soil pH, the p-values were below the test statistic (Table 5). The other explanatory variables, K, Ca, Mg, Zn, Mn, Cu, B, and N were below the respective test statics, but too near to indicate a strong statistical inference as an explanatory variable for the variance.

Many of the species were centered along the soil depth vector. However, the NTP site is a gently sloping habitat, whereas the BMT site is often a steep to nearly vertical slope. Soil depths at the BMT were often less than 1 cm. When the soil depth was removed from the CCA, elevation remained the key influence in the distribution and presence of species (Figure 16). This plot revealed that elevation was the predominant factor giving insight to the current vegetative distributions. The vegetation composition of the BRP sites were most strongly correlated with elevation, nearly falling on the elevation vector, whereas the NTP sites were strongly influenced by elevation. The statistical software identified certain environmental factors that were useless constraints. They were removed from the estimation, and no biplot scores or centroids were calculated for these constraints (R Core Team 2015).

The PCA ordination among all 12 sites showed that the presence of the *Hydatica* sp.1 was correlated with the low-elevation NTP sites and that *H. petiolaris* was correlated with the high-elevation BMT sites (Figure 17). The total variance of the species between sites is calculated, with Eigen values explaining how much variance was explained by each of the principal components axes. Vectors were drawn for species *Hydatica* sp.1 and *H. petiolaris*, indicating the direction from the origin for which sites have larger abundances for the respective species. Sites BMT 1 and BMT2 fell nearly on the *H. petiolaris* vector, whereas the NTP plots were scattered to the negative trajectory of the *H. petiolaris* vector, patterning itself around the *Hydatica* sp.1 vector.

Morphological Analysis

The morphological characteristics revealed that the BMT study sites had a zygomorphic floral symmetry, three clawed, spotted petals and two cuneate, unspotted petals (Figure 4, A), and statistically smaller petal sizes when compared to the NTP, which is radially symmetrical with all petals clawed and spotted (Figure 4, B). A principal coordinates analysis (PCoA) of 150 petal length and width measurements showed distinction between the NTP and BMT populations. The individual petal measurements grouped into clusters according to species (Figure 18). A paired t-test of the petal data also produced a p-value < 0.0001.

Phylogenetic Analysis

For taxa outside of this study, Saxifragaceae sequence data was limited to those stored in GenBank and total loci agreement between all species was not entirely possible with only three loci available among the major genera. Multiple sequences were downloaded for each species identified and concatenated to develop at least one representative of each genus within the study. A total of 101 sequences were initially developed for the ITS1, 5.8s, ITS2 regions and processed for phylogenetic inference. The resulting topology represented a worldwide (global) representation of familial clades within the Saxifragaceae. The genera were then reduced to 77 sequences to allow for those found throughout the BRE, representing 11 out of 25 genera for the family Saxifragaceae, with a total of 20 alignments used for final topologies. A total of 645 base pairs (bp) were extracted from the original 998 bp through GBLOCKS, to represent 65% of the original alignment performed in MUSCLE. The removal of poorly aligned regions was relaxed when compared to a strict analysis performed in GBLOCKS, which retained only 36 bp (4%) of the original 998 bp. The relaxing of the selection of poorly aligned regions allowed smaller final blocks, gap positions within the final blocks, and less strict flanking positions.

The concatenated molecular portion that resulted had 292 characters that were parsimony informative (69.7%), with a consistency index of 0.21 and a retention index of 0.62. The topology was similar between Maximum Parsimony (MP) and Maximum-Likelihood (ML) analysis, and initial ML in SATe showed clear distinction for *Hydatica* sp.1 (Liu et al. 2011) (Figure 19).

Within the global phylogeny, the resulting alignments agreed with phylogenetic work by Deng, et al. (2015), on the Saxifragaceae (Figure 20). The NCBI Blast search developed an initial phylogeny based on pairwise alignment methodology that excluded multiple alignment, which placed the NTP population as a sister taxon to *Hydatica petiolaris* (Figure 21). Eleven of the 15 genera were strongly supported as clades across analyses. Analysis of the Maximum Likelihood in both SATe and MEGA 6.0 resulted in a phylogeny that provided a bootstrap score of 97% (Figure 22).

The BRE phylogeny was similar in clade agreement with the global phylogeny. The NCBI Blast search for the BRE individuals also produced an intial phylogeny based on pairwise alignments, which placed the NTP population as a sister taxon to *Hydatica petiolaris* (Figure 23). A final BEAST analysis, which incorporated the MCMC analysis produced a posterior probability of 95%, again placing *Hydatica* sp.1 as a sister taxon to *Hydatica petiolaris* (Figure 24).

CHAPTER 4

DISCUSSION & CONCLUSION

Analysis indicates that there is enough heuristic evidence supporting *Hydatica* sp.1 as a novel species under the Biological Species Concept (BSC), Ecological Species Concept (ESC), and Phylogenetic Species Concept (PSC), thus requiring a taxonomic reevaluation of *Hydatica petiolaris* (Raf.) Small, *sensu lato*.

Within the *Hydatica* Group, *Hydatica* sp.1 has a very close morphological resemblance to *H. petiolaris* but is distinct based on several features, including floral arrangement, nectary spots, and petal morphology. The flowers of *Hydatica* sp.1 are similar to those of *H. petiolaris* in that petals of both are distinctly clawed but *Hydatica* sp.1has 5 clawed petals, whereas *H. petiolaris* has 3 clawed petals and two cuneate petals. The petals of *Hydatica* sp.1 each have 2 nectary spots adjacent to the claws of each petal, whereas *H. petiolaris* has 2 spots only on the upper three petals, with the two lower, cuneate petals lacking nectary spots. The two species are also distinct at the molecular level, with differences in nuclear ITS (a fixed difference at multiple positions). The phylogenetic analysis is consistent with previous studies in supporting the division of Saxifragaceae into the *Micranthes*, *Hydatica*, and *Saxifraga sensu stricto* (Soltis et al. 1996, 2001).

Species Concept Support

Criteria for a population or individual to meet the BSC were originally outlined by Ernst Mayr, in that species are “groups of actually or potentially interbreeding natural populations, which are reproductively isolated from other such groups" (Mayr 1942, 1963). The observation of the NTP *Hydatica* sp.1 population as flowering and setting fruit at a segregated time period (late February to early May) from the BMT *Hydatica petiolaris* population (late May/early June to September), both in the open garden experimentsand the field, meets this criterion. Segregated flowering are shown as indicators to historical speciation events in a diverse range of taxa, in association with different pollinator attraction or ecological stresses (Macnair, et al. 1989; Gottleib 1979; Kipling and Warren 2014).

Separate floral morphology and flowering times also indicates a possible shift in pollination strategy indicative of a historic speciation event (Aceto et al. 1999; Gregg 1983; Johnson 1996). These shifts follow an evolutionary trend among vascular flora, transforming from out-crossing individuals to selfing populations or vice-versa, and are usually coupled with morphological changes (Crawford et al. 2014). The petal data reveal a difference between the NTP and BMT populations, in that the NTP population have 5 clawed petals that are predominantly larger in dimension than the BMT petals, and arranged in an actinomorphic, radial pattern. Neal et al. (1998) and Endress (2001) establish the presence of two or more `symmetry planes' to define actinomorphy, whereas zygomorphy is along only one plane. The shift in floral arrangement has been noted as a strategy for both attracting pollinators and indicating presence of nectar rewards (Rudall and Bateman 2002). The PCoA ordination graph identified the variation between the NTP and BMT petal measurements, forming clusters and identifying the two species as independent in petal size, in addition to phenology and floral symmetry. The distinctiveness of *Hydatica* sp.1 from *H. petiolaris* is also supported by the paired t-test which produced a p-value < 0.0001, and as such the two species are more ordered as distinct species than would be expected by chance.

Under the ESC, a species or lineage has to occupy a different niche than sister taxa to be considered a monotypic taxon (Van Valen 1976; Andersson 1990). *Hydatica petiolaris* and *Hydatica* sp.1 do occupy their own ecological niche. The statistical ordination graphs depict the ecological and community survey data as distinctly segregated to the NTP and BMT sites. The PCA ordination of the ecological data reflected that among the composition of the 12 surveyed sites, *Hydatica* sp.1 was endemic to the NTP site. The soils data also provides some insight for the *Hydatica* sp.1 and *H. petiolaris* community types, in that these species may be segregated to a high magnesium or calcium site throughout the BRE (Proctor 1971).

Weather data also provide evidence of differences between the two sites, often demonstrating that the higher elevation BMT site had colder temperatures throughout the year than the NTP site. Both the macro- and micro-climatic data reveal statistical evidence of temperature variations between the two sites in reference to average daily and monthly temperatures. The ANOVA tests reveal a p-value < 0.0001 between the data from both sites, further strengthening the findings of the CVS survey and soils data.

The PSC is uniformly accepted as being a concept supported by a diagnosable cluster of individuals within which there is a parental pattern of ancestry and descent among units of like kind (Eldredge and Cracraft 1980). It is often used as the standard for DNA barcoding and taxonomy studies (Kelly et al. 2007). For the PSC, the strongest support for monophyletic distinction resulted from the Bayesian Evolutionary Analysis by Sampling Trees (BEAST) analysis, which places the NTP population of *Hydatica* sp.1 separate from the BMT populations of *Hydatica petiolaris* .

CHAPTER FIVE

TAXONOMIC TREATMENT AND KEY

Given the aforementioned evidence, I present herein a proposed taxonomic treatment, diagnosis, description, distribution, and taxonomic key.

Hydatica sp.1., is described from a low elevation granite dome, in Pickens County, SC. It is clearly closely related to *H. petiolaris* (Raf.) Small but differs in its annual habit and floral morphology.

TYPE: **UNITED STATES. South Carolina.** Pickens County, northeast city of Pickens, along edge of open granite outcrop of Cedar Rock Mountain, just S of East Preston McDaniel Road and W of Nine Times Road, 34.946423N, -82.802374W, 438.9 m. Plants most abundant at edge of woodland, under cedar trees, on mossy mats, first observed in 2002, (holotype: CLEMS).

Annual herbs, stems 1-5 dm high, rising from basal rosettes, glabrous but usually finely and moderately puberlant. **Leaves** oblanceolate, to 15 cm long and 4 cm wide, acute, coarsely toothed, base attenuate, arranged in rosette. **Peduncle** glabrous but usually finely and moderately puberlant with diminutive leaves scatted alternately along scape. **Flowers** in diffuse panicle**. Calyces** 1.5-2.5 mm long; sepal lobes lanceolate, reflexed in fruit. **Petals** 3-4 mm long, each distinctly clawed (petal blade with cordate or truncate base) and with 2 yellow spots. **Stamens** 10, barely exserted, filaments filiform, anthers yellow, ca 0.5 mm long. **Ovary** ca 3.5-7 mm long, ovoid, superior, nerved into two halves, slowly tapered into styles. **Styles** 0.5-1mm long. **Seeds** longitudinally striate, echinate, 0.8-1 mm long.

Similar to a depauperate *Hydatica petiolaris* (Raf.) Small except for the annual habit, much earlier flowering, and smaller flowers that are only weakly zygomorphic and more frequently radially symmetrical.

According to Radford et al. (1968) and Weakley (2012), *Hydatica petiolaris* has a corolla that is bilaterally symmetrical, the 3 upper petals distinctly clawed (the petal blade with a cordate or truncate base) with 2 yellow spots and the 2 lower petals smaller, cuneate, and not spotted; leaf margins coarsely dentate. Our examination of *Hydatica* sp.1 showed that the corolla is radially symmetrical, spotted, and clawed. This is the only known Blue Ridge Escarpment population of *Hydatica* sp. at a low elevation with this unique morphology.

SAXIFRAGACEAE A.L. de Jussieu 1789 (Saxifrage Family)

A family of about 35 genera and 500-650 species, herbs (mainly perennial), nearly cosmopolitan, but especially diverse in warm temperate and cold temperate regions of North America and Eurasia. The circumscription of a much narrower Saxifragaceae is clearly warranted, based on a wide variety of data, and recently strongly corroborated by molecular data (Soltis et al. 1993) (Weakley 2012)

1. Leaves compound .... ***Astilbe***

1' Leaves simple (sometimes cleft or lobed).

2. Stem creeping, the leaves all cauline, opposite, short-petioled or sessile, < 2 cm long ....***Chrysosplenium***

2' Stem erect, the leaves mostly or entirely basal, alternate (stem leaves opposite in *Mitella*), long-petioled, > 4 cm long (except short petioled or sessile and sometimes < 4 cm long in *Micranthes*).

3. Basal leaves short-petioled or sessile, petioles 0-1× as long as blade; basal leaves cuneate or rounded at the base; leaf venation predominately pinnate.

4. Corolla bilaterally symmetrical or radially symmetrical, some petals with 2 yellow spots, clawed and/or cuneate; leaf margins coarsely dentate ....***Hydatica***

4' Corolla radially symmetrical with no spots; leaf margins entire to serrate ... ***Micranthes***

3' Basal leaves long-petioled, petioles (1-) 2-5× as long as blade; basal leaves cordate at base; leaf venation predominantly palmate.

5 Stem leaves opposite; petals fimbriate; inflorescence a raceme; flowers on pedicels 1.5-3 mm long .... ***Mitella***

5' Stem leaves absent or alternate; petals not fimbriate; inflorescence a panicle or raceme; flowers mostly on pedicels > 3 mm long.

6 Inflorescence racemose; stamens 10 .... ***Tiarella***

6' Inflorescence paniculate; stamens 5.

7 Seeds winged, 1.3-1.5 mm long; leaves cleft < ½ way to base; hypanthium fused to pistils only at bases; stems
normally with several petiolate leaves much like basal leaves (though typically somewhat smaller) .... ***Sullivantia***

7' Seeds papillose, echinate, smooth, or slightly ridged, 0.4-0.7 mm long; leaves cleft > ½ way to base (in *Boykinia*) or < ½ way (in *Heuchera*); hypanthium fused to lower half or more of pistils; stems with (in *Boykinia*) or without (in *Heuchera*) several petiolate leaves.

8 Stems normally with several petiolate leaves much like the basal leaves (though typically somewhat smaller); ovary with 2 locules; leaves cleft > ½ way to base ....***Boykinia***

8' Stems with only reduced sessile bracts unlike basal leaves; ovary with 1 locule; leaves cleft < ½ way to base ....***Heuchera***

Hydatica Necker ex Gray 1821 (Appalachian Saxifrage)

A genus of about 12 species, herbs, of temperate W. North America, Europe, and E. North America. In crevices in exposed rock outcrops at high-elevations, other rock outcrops (moist to rather dry), periglacial boulderfields, rocky seeps, often exposed but sometimes shaded. February-August. Leaves mostly with 4-8 teeth per side; pubescence of the leaves and scapes mostly gland-tipped; corolla bilaterally symmetrical or radially asymmetrical; filaments filiform; [mostly of rock outcrops and seepages, often exposed, but sometimes shaded The orange anthers are an attractive contrast to the white petals (some with the three upper bearing two yellow spots each or all with two yellow spots each). A Southern Appalachian endemic: northwest VA, WV, and KY south to eastern TN, w. NC, southwestern SC, and northeastern. GA.

1. Corolla bilaterally symmetrical, 3 upper petals distinctly clawed (petal blade with a cordate or truncate base) and with 2 yellow spots, the 2 lower petals smaller, cuneate, and not spotted; filaments filiform .... ***H. petiolaris***

1' Corolla radially asymmetrical, each petal distinctly clawed and with 2 yellow spots; filaments filiform; [mostly of rock outcrops and seepages, often exposed, but sometimes shaded) .... ***Hydatica* sp.1**

TABLES

Table 1. List of vouchers examined from the Clemson University Herbarium (CLEMS).

Species	CLEMS Accession #	Location
Saxifraga micheauxii	53193	Tallulah Gorge, GA
S micheauxii	43483	Big Creek Trail, GA
S micheauxii	54386	Blood Mountain, GA
S micheauxii	24043	Glade mountain, NC
S micheauxii	88444	Chestnut Bald, NC
S micheauxii	To Be Accessioned (TBA)	Balsam Mountain Region, NC
Hydatica petiolaris	TBA	Cedar Top Mtn , SC

Table 2. First open flower months of NTP and BMT open garden plots vs NTP and BMT field study plots.

2013 & 2014 Observations of first flowering					
Site	BM	TG	BCT	BMT	NTP
Open Garden	June	June	July	May	February
Field Site	June	June	July	May	February

Table 3. Summary table of NTP and BMT monthly weather data segregated into High, Low, and Average temperatures, with ANOVA of NTP and BMT monthly weather data following.

Monthly Temperature Data (*F)

Month	HOBO CT High	BMT High	CT Low	BMT Low	CT Avg	BMT Avg
Jan-14	76 62	79 41	9 88	-1 77	35 76204854	30 16912162
Feb-14	85 1	66 96	25 93	18 81	45 02936756	37 00321429
Mar-14	80 11	95 49	23 32	11 97	48 23846774	42 11576613
Apr-14	86 56	86 56	30 09	21 54	60 04440278	48 78759028
May-14	100 18	83 66	44 65	34 88	68 21802419	56 90418011
Jun-14	104 23	80 82	60 8	49 68	74 19283046	63 03052443
Jul-14	101 79	79 41	60 12	48 25	74 71990591	62 54609543
Aug-14	93 22	100 18	62 17	48 96	73 96623237	64 48602418
Sep-14	88 74	103 41	53 19	43 92	69 61675	62 678
Oct-14	80 11	101 79	34 88	28 45	60 51725806	54 57176747
Nov-14	75 22	98 6	20 64	11 97	46 78831367	44 28652325
Dec-14	73 15	92 46	29 28	25 07	46 63615591	43 14063844
Jan-15	63 54	77 31	9 88	-2 27	38 21660787	37 21079838

Single Factor Anova

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
HOBO CT High	13	1108 57	85 27461538	148 3166436
BMT High	13	1146 06	88 15846154	128 4171141
CT Low	13	464 83	35 75615385	349 5996256
BMT Low	13	339 46	26 11230769	335 4405026
CT Avg	13	741 9463651	57 07279731	203 3044388
BMT Avg	13	646 930244	49 76386492	136 2240945

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	41964 23274	5	8392 846548	38 69744538	4 28425E-19	2 341827531

Within Groups	15615 62903	72	216 8837365
Total	57579 86177	77	

Table 4. Rank Abundance tables with top ten rankings of CVS survey depicting community composition

Ntp Site	Rank	Abundance (%)	Proportion	Plower	Pupper	Accumfreq
Rock	1	33	7.2	3.3	11.1	7.2
Pinus virginiana	2	20	4.4	0.1	8.7	11.6
Juniperus virginiana	3	18	3.9	1.8	6.1	15.6
Polytrichum commune	4	17	3.7	1.7	5.7	19.3
Misc. Moss	5	16	3.5	0.3	6.7	22.8
Yucca smalliana	6	16	3.5	0.3	6.7	26.3
Misc. Lichen	7	15	3.3	0.2	6.4	29.6
Grimmia sp.	8	14	3.1	1.0	5.2	32.7
Selaginella tortipila	9	14	3.1	-0.5	6.6	35.7
Minuartia glabra	10	10	2.2	-0.5	4.9	37.9

Bmt Site	Rank	Abundance (%)	Proportion	Plower	Pupper	Accumfreq
Rock	1	14	9.7	2.9	16.6	9.7
Sphagnum sp.	2	10	6.9	2.0	11.8	16.7
Cyperus sp.	3	9	6.2	-7.0	19.5	22.9
Cladonia caroliniana	4	8	5.6	-25.8	36.9	28.5
Grimmia sp	5	7	4.9	-0.5	10.3	33.3
Rubus allegheniensis	6	7	4.9	-25	34.8	38.2
Erigeron sp	7	6	4.2	1.2	7.1	42.4
Rhododendron vaseyii	8	6	4.2	-10.5	18.9	46.5
Solidago sp	9	6	4.2	-10.5	18.9	50.7
Hydatica petiolaris	10	5	3.5	-2.9	9.8	54.2

Table 5. ANOVA testing of environmental explanatory variables used in CCA analysis.

Explanatory variable	Response Variables					
	Hydatica sp 1			Hydatica petiolaris		
	F	P	% of expl Var	F	P	% of expl Var
soilDepths	30.036	0.03172*	93.8	3.41E+10	2.935E-11***	100.0
elevation	1299.7	.0007685***	99.8	3.65E+10	2.736E-11***	100.0
flowering time	1.67E+10	5.971E-11***	100.0	3.65E+10	2.735E-11***	100.0
SoilpH	0.1281	0.7546	6.0	0.2645	0.6583	11.7
P	3.0922	0.2207	60.7	4.75E+10	2.103E-11***	100.0
K	72.825	.01345*	97.3	3.60E+10	2.977E-11***	100.0
Ca	1.6844	0.3239	45.7	5.96E+10	1.678E-11***	100.0
Mg	0.0522	0.8406	2.5	0.0943	0.7878	4.5
Zn	41.929	.02303*	95.4	3.54E+10	2.824E-11***	100.0
Mn	0.8903	0.445	30.8	5.22E+10	1.916E-11***	100.0
Cu	4.8065	0.1597	70.6	7.32E+10	1.366E-11***	100.0
B	2.0118	0.2919	50.1	3.37E+10	2.965E-11***	100.0
Na	0.0269	0.8848	1.3	0.0486	0.8461	2.4

*Signif codes: '***' 0 001 '**' 0 01 '*' 0 05 ' ' 0 1 ' ' 1*

FIGURES

Figure 1. The Blue Ridge Escarpment traversing northeastern Georgia through South and North Carolina, into Virginia.

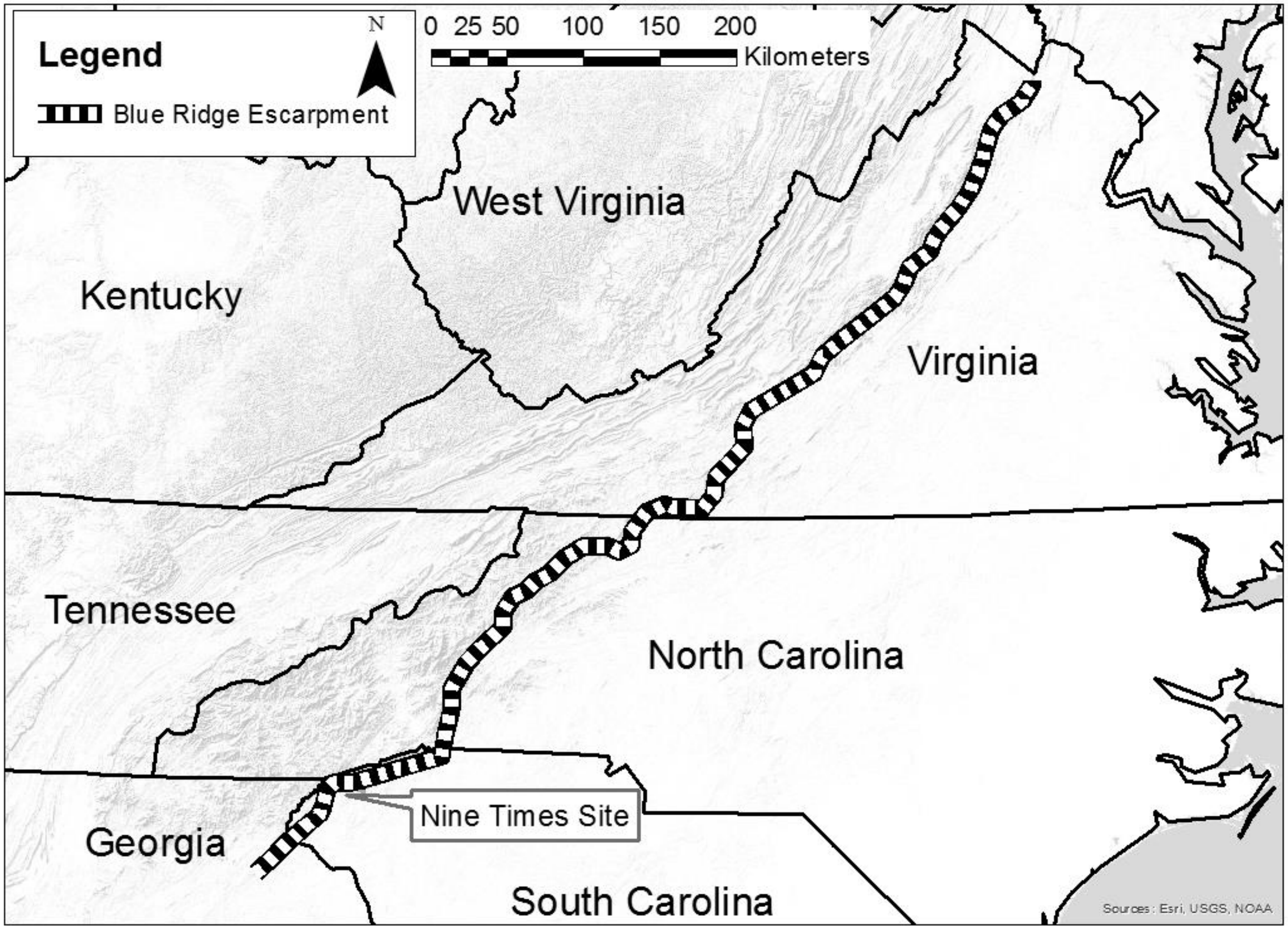


Figure 2. The Nine Times Nature Conservancy Site, located in Pickens, County, SC.

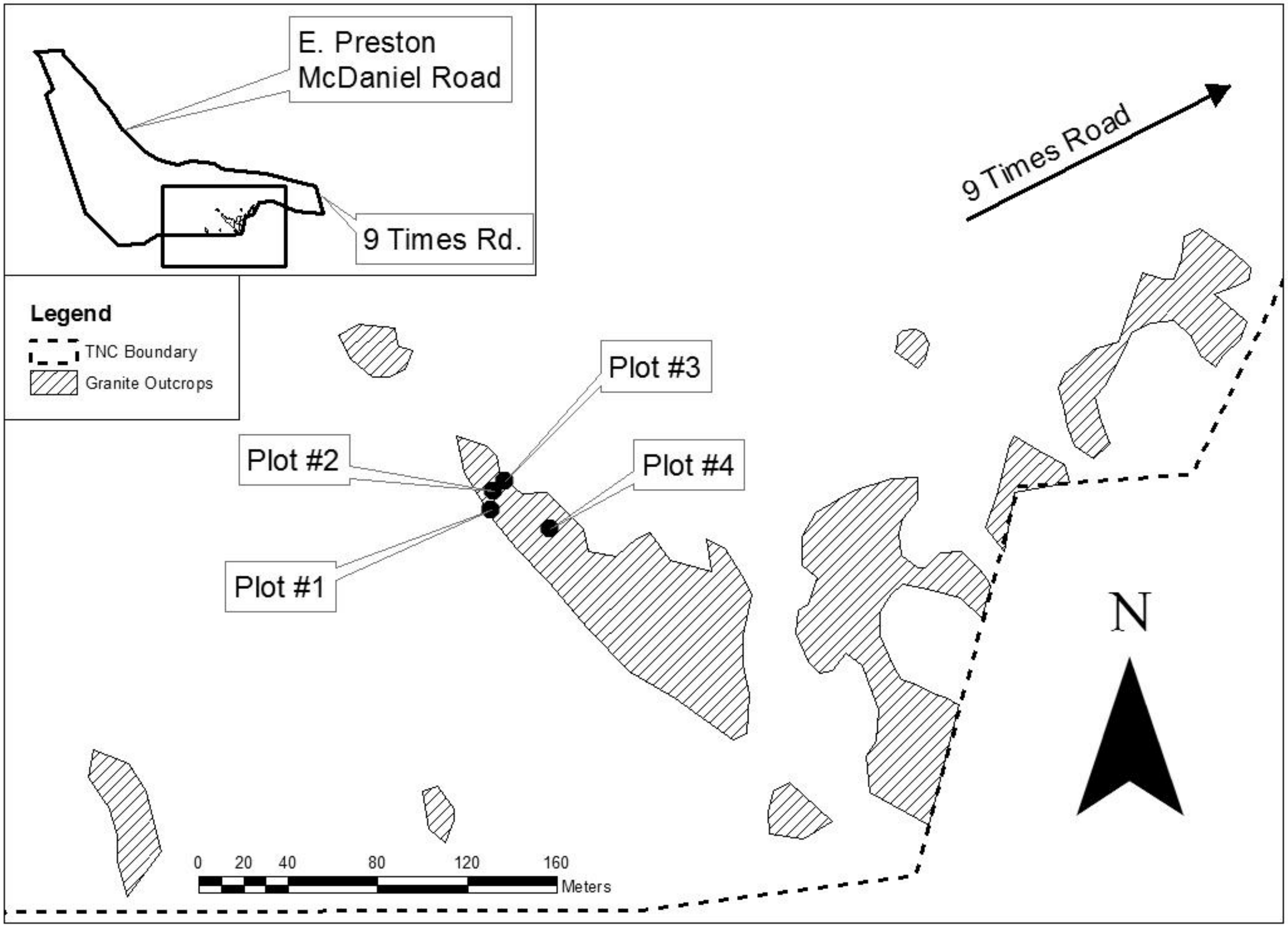


Figure 3. Representatives of the family *Saxifragaceae* depicting variable floral morphology and arrangements.A, *Boykinia*, B, *Chrysosplenium*, C, *Micranthes*, D, *Ozomelis*.

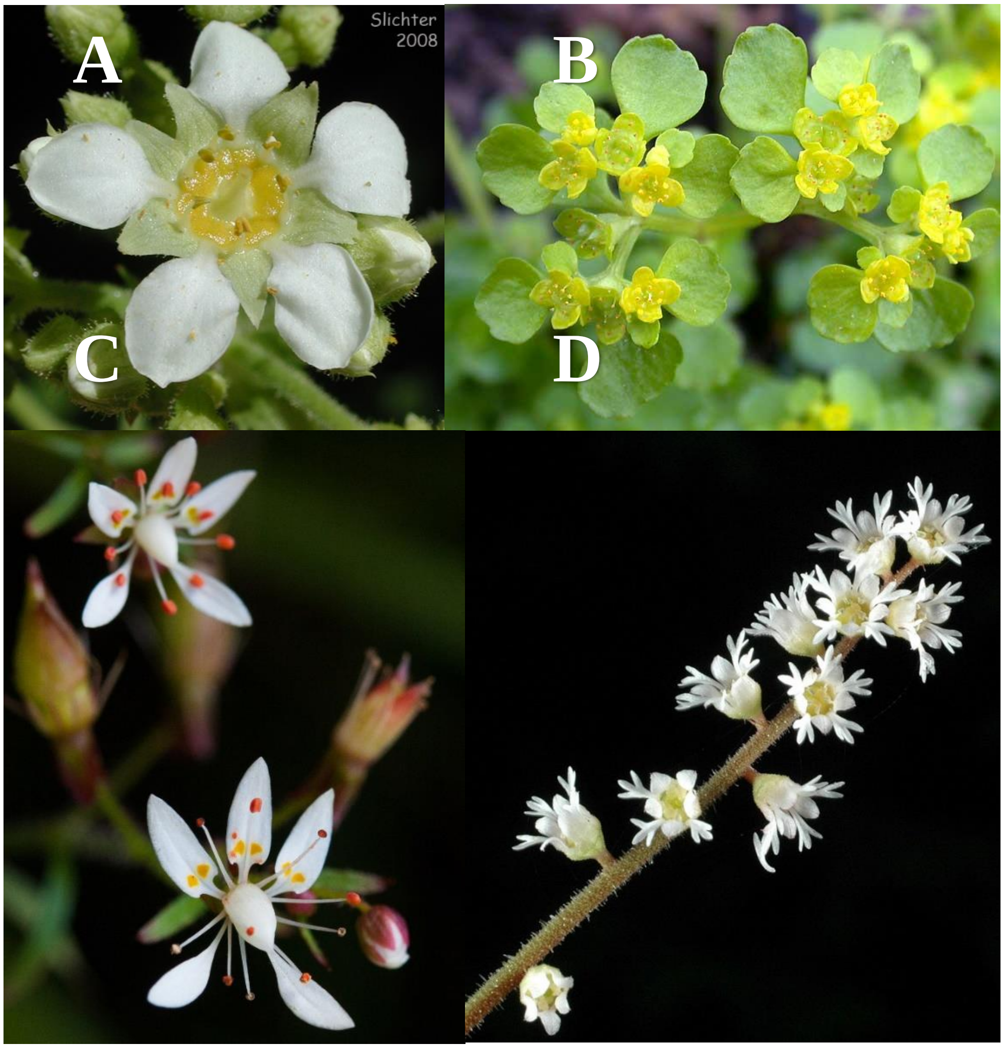


Figure 4. A) High-elevation *Hydatica petiolaris* B) low-elevation *Hydatica* sp.1.

Figure 5. Study site locations: Nine Times Preserve (NTP), SC, 440 meters (m) above sea level (ASL); Blood Mountain (BM), GA 1036m ASL; Tallulah Gorge (TG), GA, 428m ASL; Big Creek Trail (BCT), GA, 670m ASL; and the Balsam Mountains region (BMT), NC 1584m ASL.

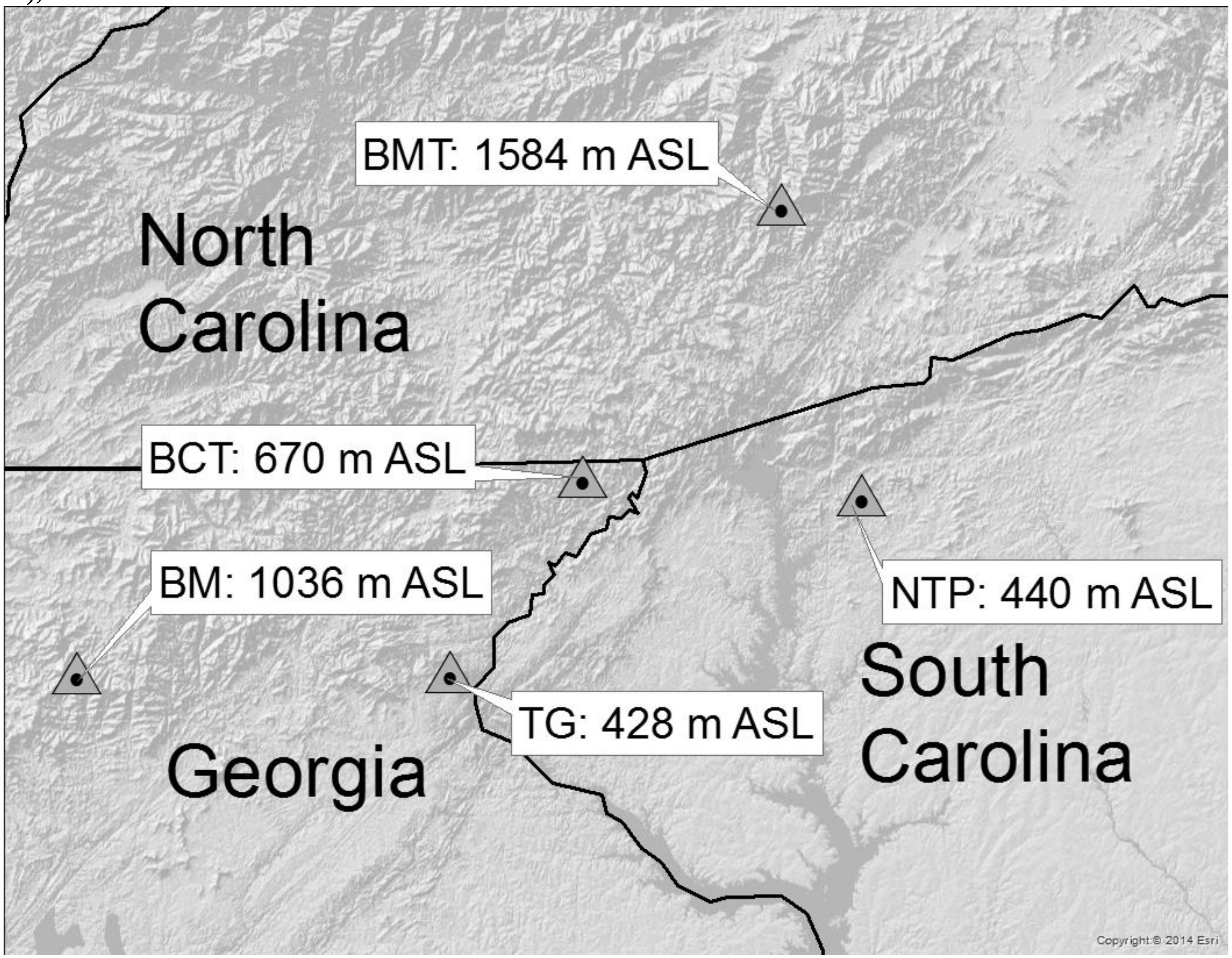


Figure 6. Typical $1m^2$ study plot at the NTP study site. The *Hydatica* sp.1 population is in flower.

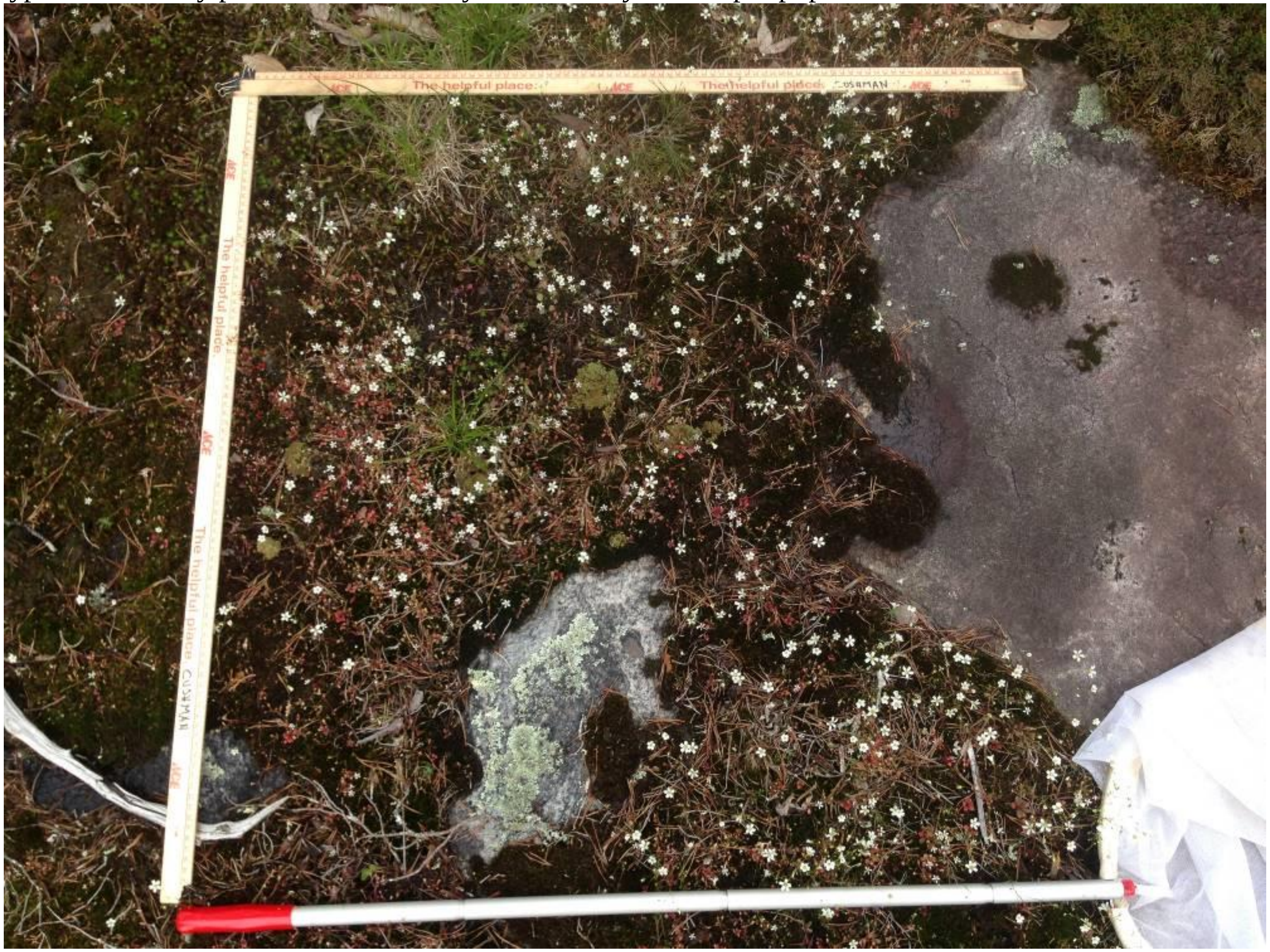


Figure 7. Hydatica sp.1 petals mounted on slides for measurement of length and width data with microscope.

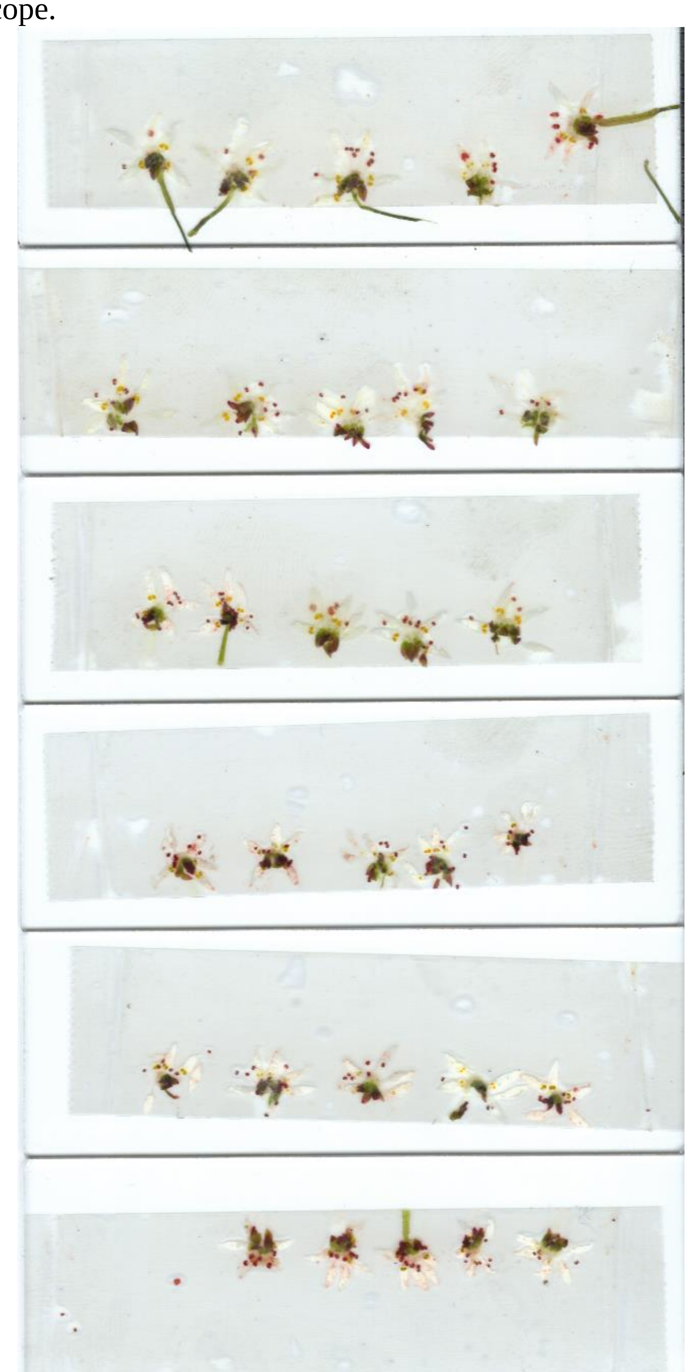


Figure 8. Controlled Garden Experiment plantings at the South Carolina Botanical Garden greenhouse. Ambient garden planting trays shown with orange tags representing 2013 plantings, white tags representing 2014 plantings.

Figure 9. Patrick D. McMillan performing portion of vegetative survey at NTP. Author is taking picture.

Figure 10. Comparative weather data of the NTP and BMT sites for study cycle years 2014 – 2015. NTP and BMT flowering start times indicated by shaded bars.

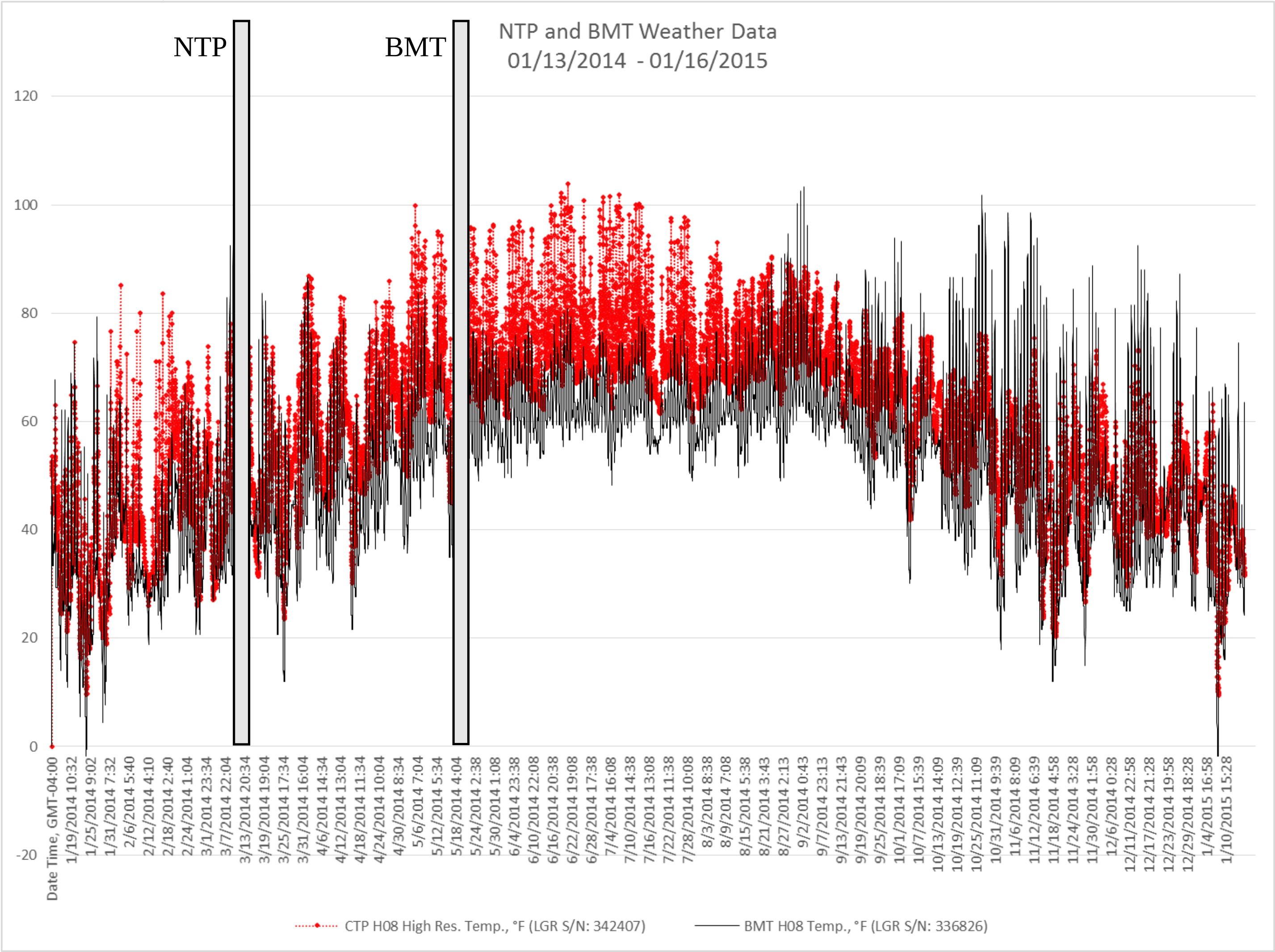


Figure 11. National Weather Service's National Digital Forecast Database (NDFD) Annual weather data for Pickens, SC. Range is 01/13/2014 – 01/16/2015

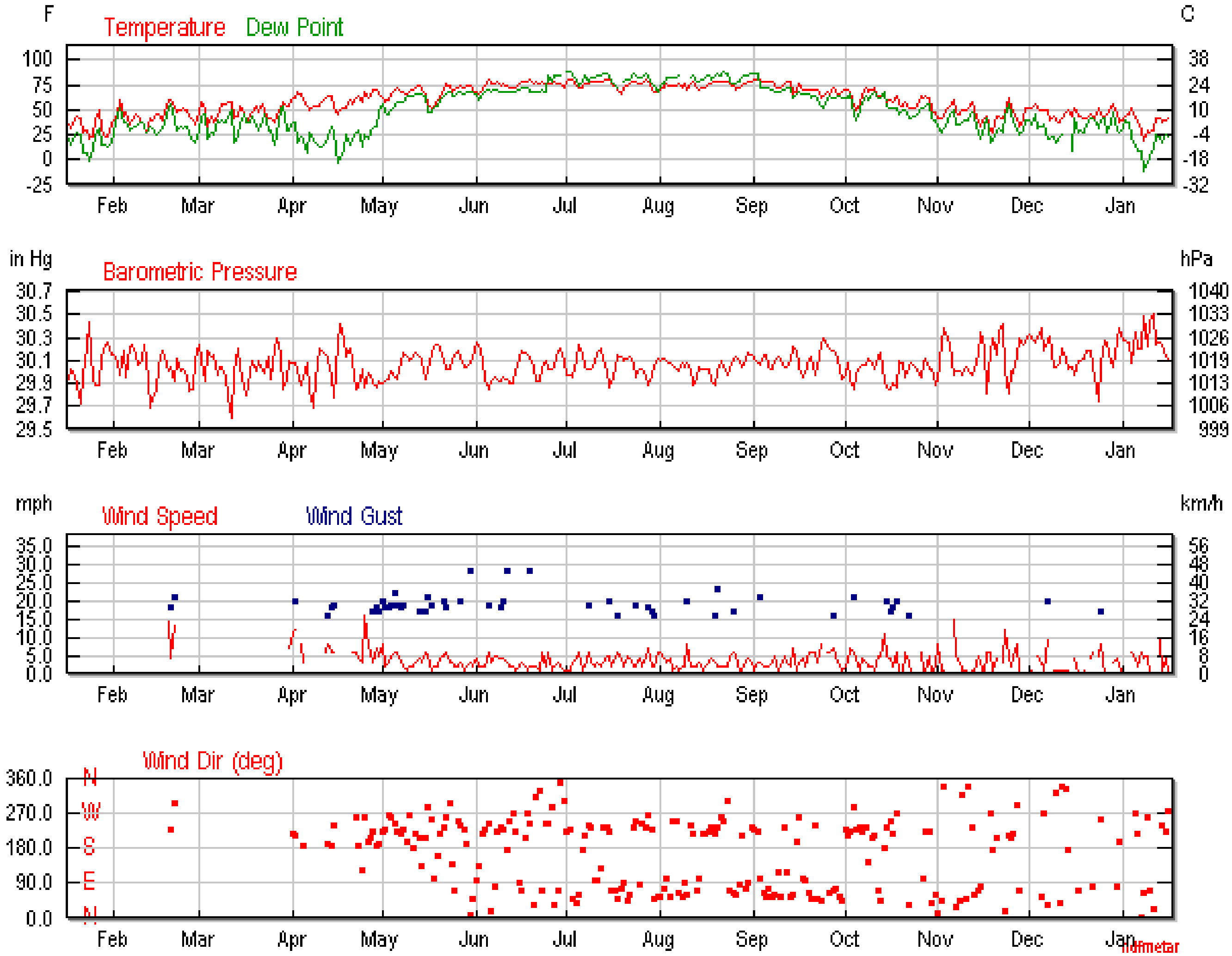


Figure 12. National Weather Service's National Digital Forecast Database (NDFD) Annual weather data for Pickens, SC. Range is 01/13/2014 – 01/16/2015

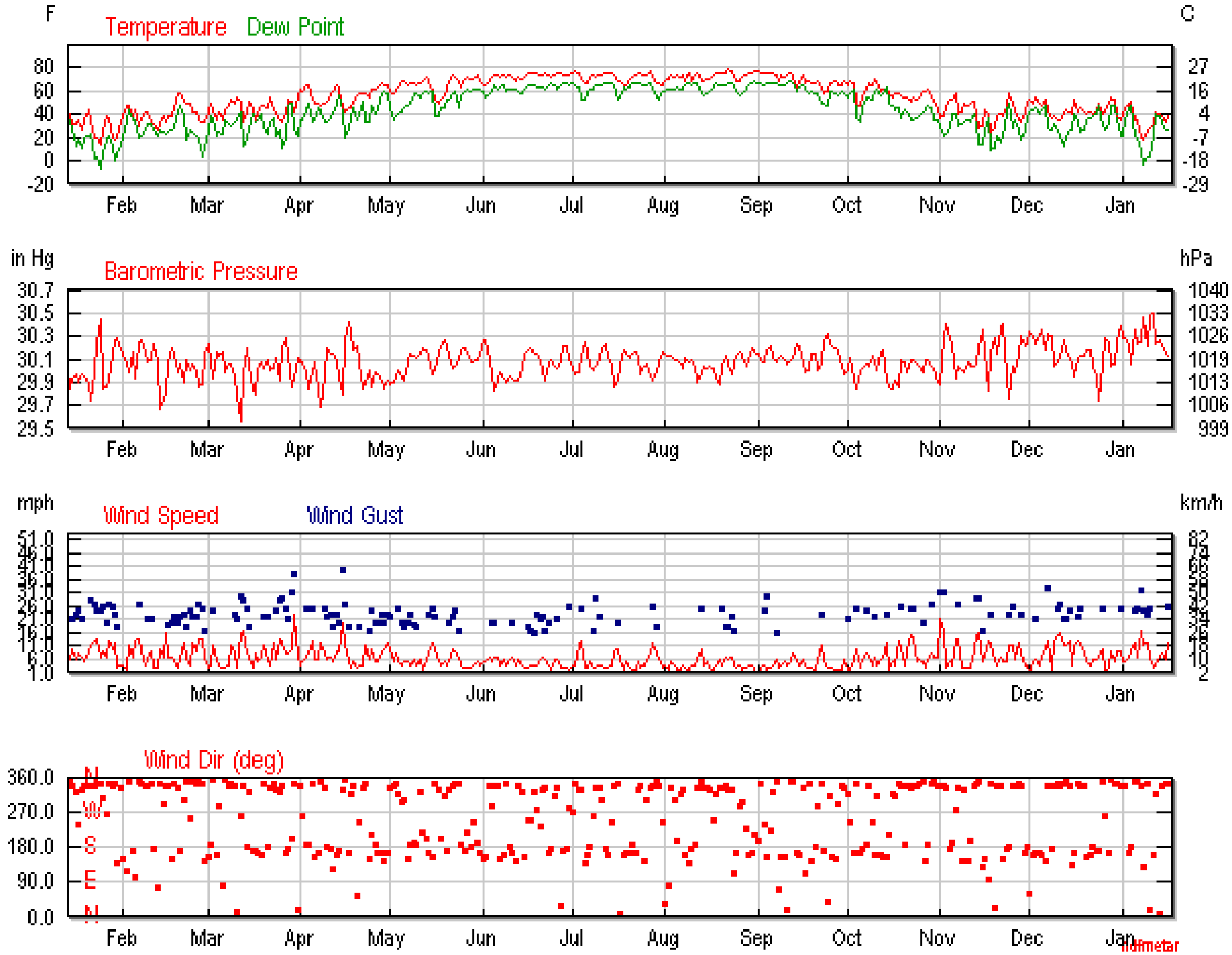


Figure 13. Rank abundance profiles of vegetative survey at NTP site.

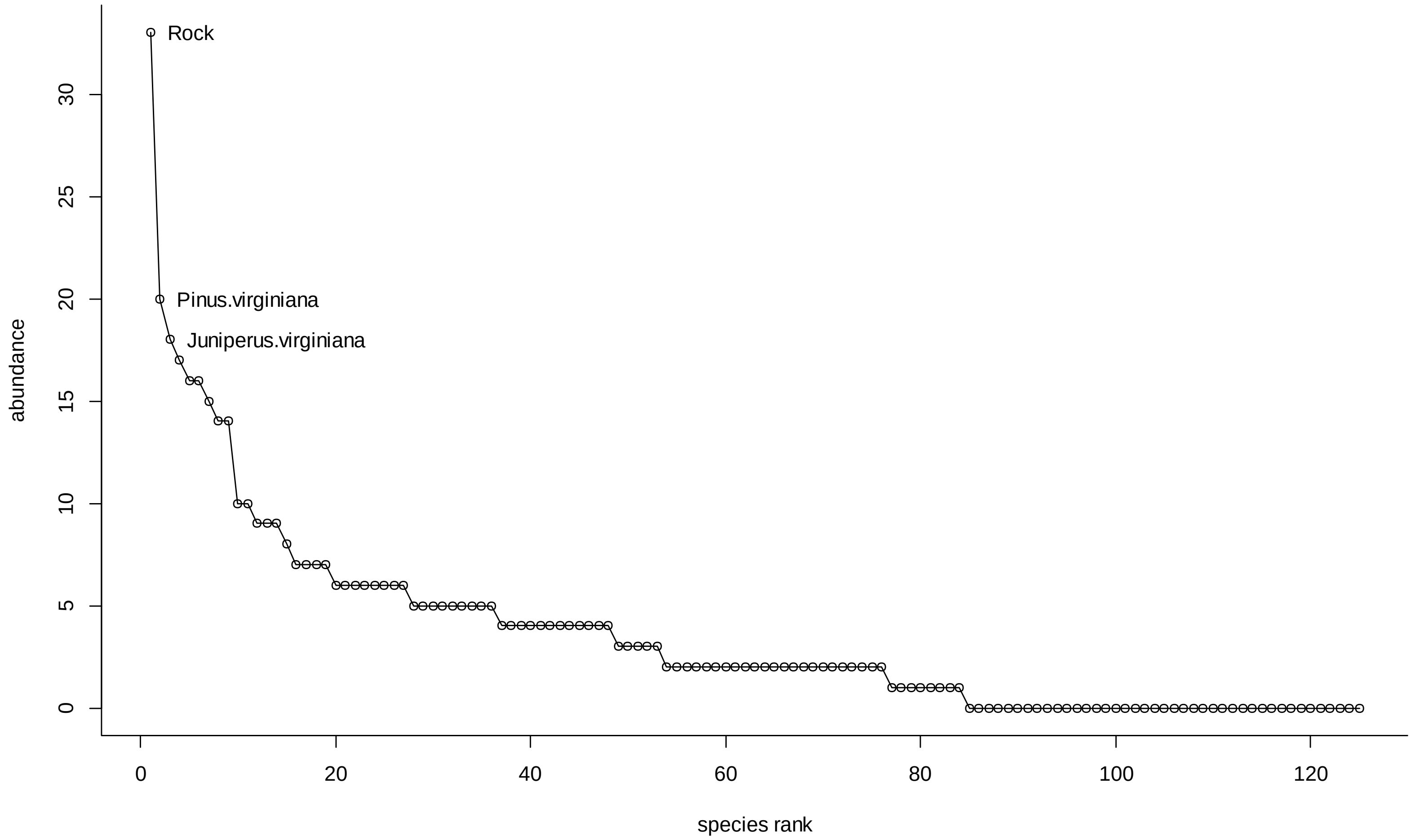


Figure 14. Rank abundance profiles of vegetative survey at BMT site

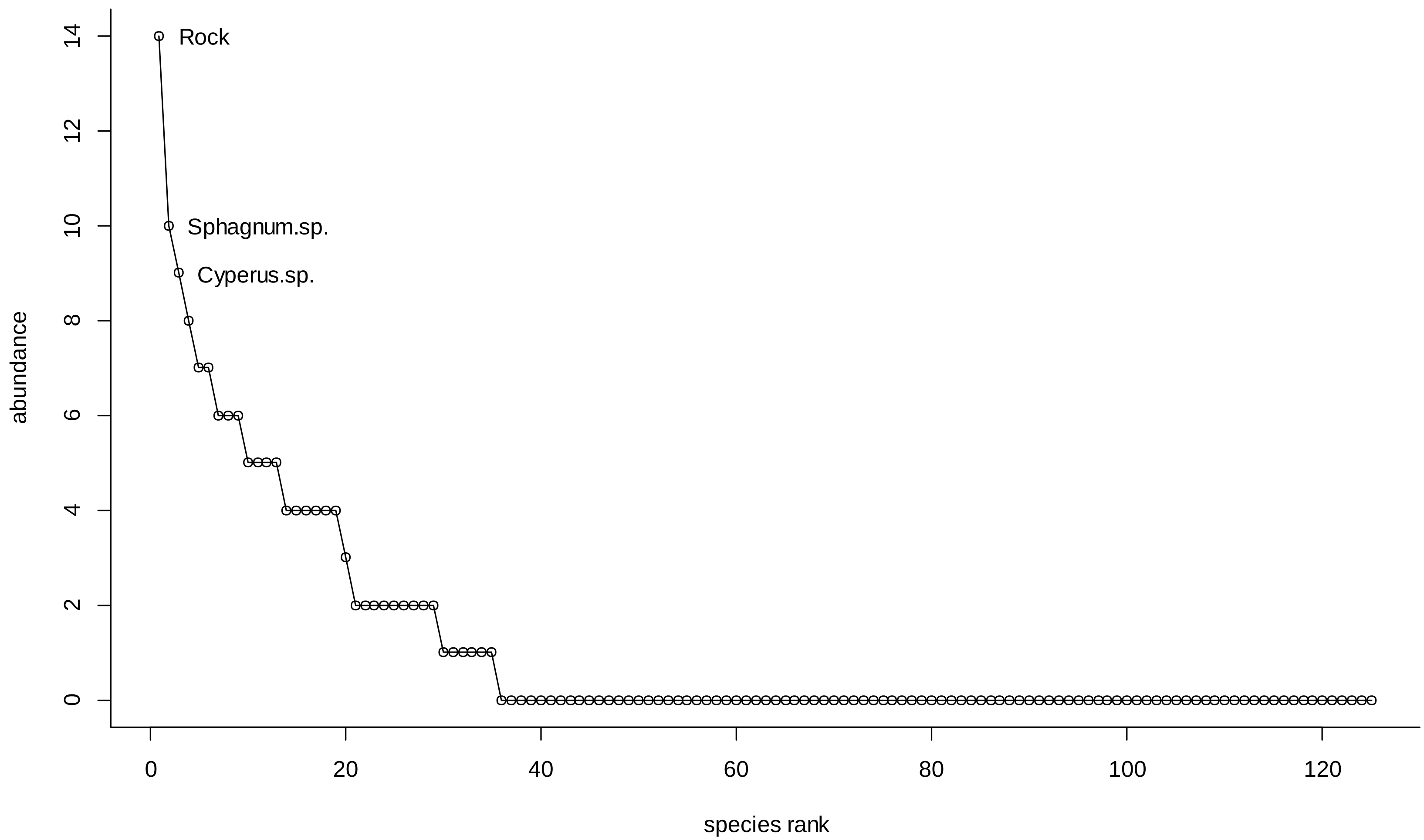
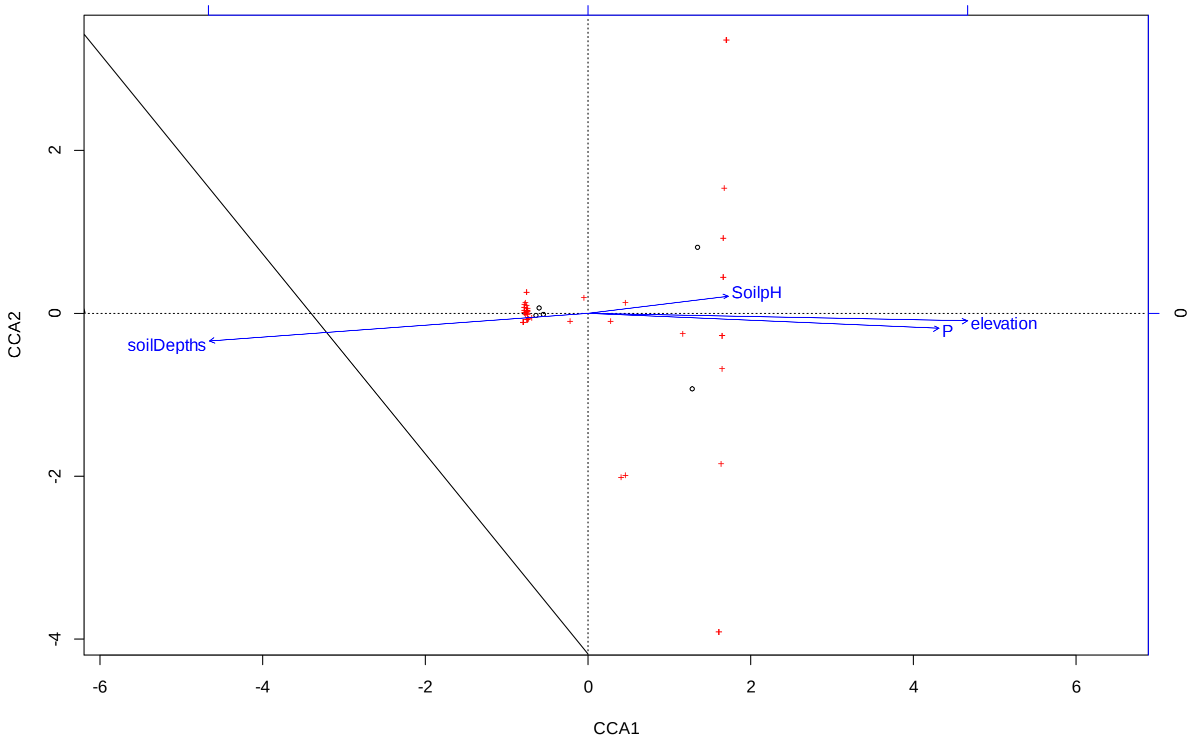
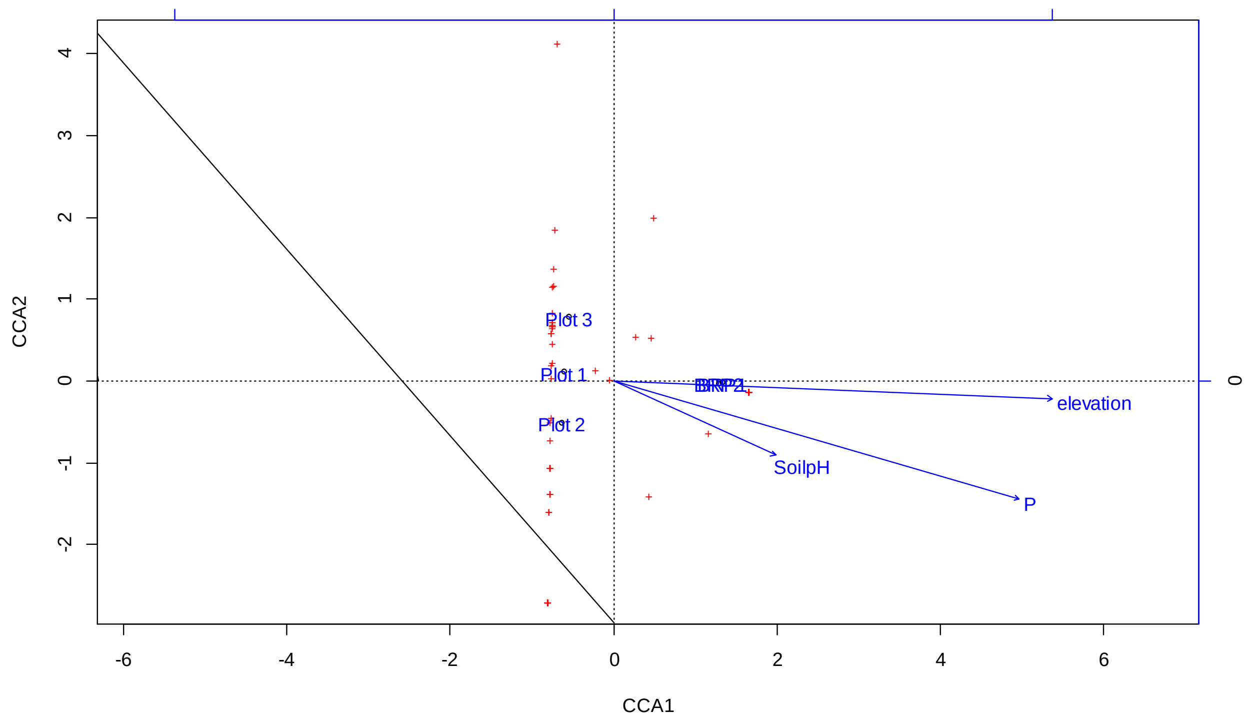


Figure 15. This plot shows us that soil depths has a great influence on the presence of species, and that the elevation is the second factor that explains distributions. Many of the species distributions are centered near the soil depths vector. Some environmental factors were also nulled due to identification as useless constraints. They are completely removed from the estimation, and no biplot scores or centroids are calculated for these constraints. (R Core Team 2015).



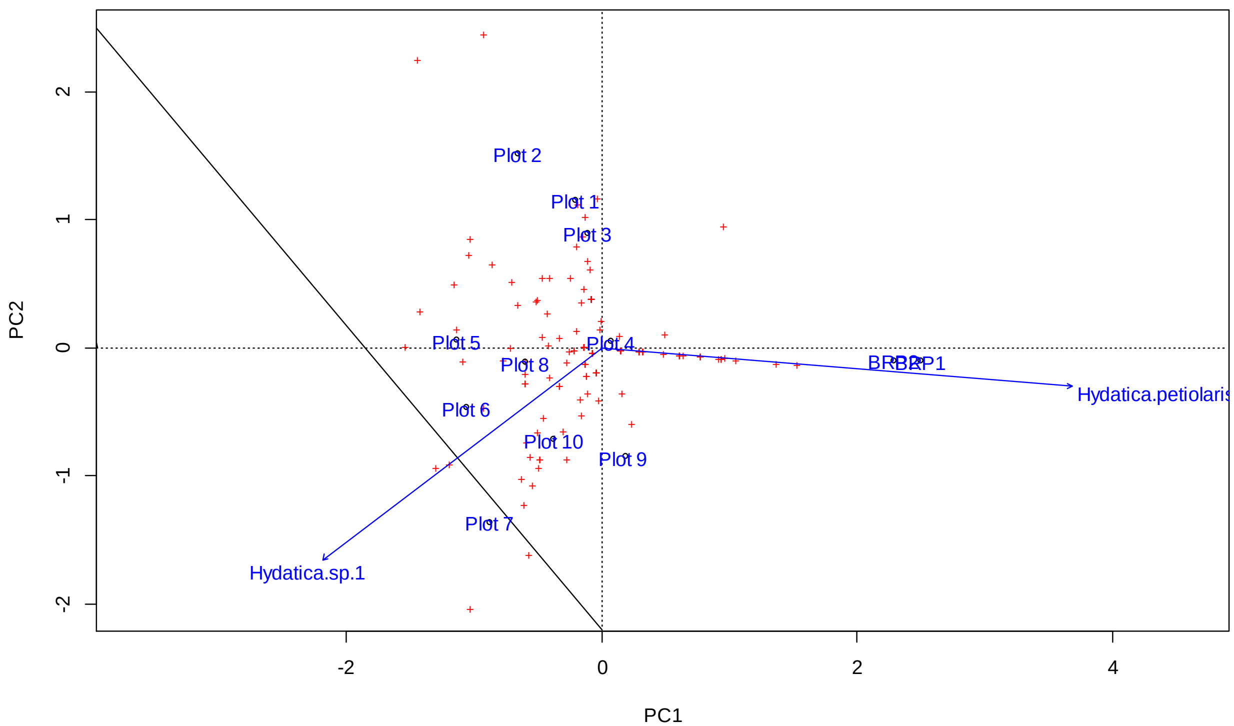
	CCA1	CCA2	CCA3	CCA4	
Eigenvalue	0.7928	0.2399	0.1894	0.1429	0
Proportion Explained	0.5808	0.1757	0.1388	0.1047	0
Cumulative Proportion	0.5808	0.7565	0.8953	1.0000	1

Figure 16. This plot shows us that the elevation is the greatest factor that explains distributions. A lot of the species distributions are centered near the soil depths vector. Also note that some environmental factors were nulled due to identification of useless constraints. They are completely removed from the estimation, and no biplot scores or centroids are calculated for these constraints. (R Core Team 2015). Eigen value for Axis 1, 0.7926, Axis 2, 0.1894.



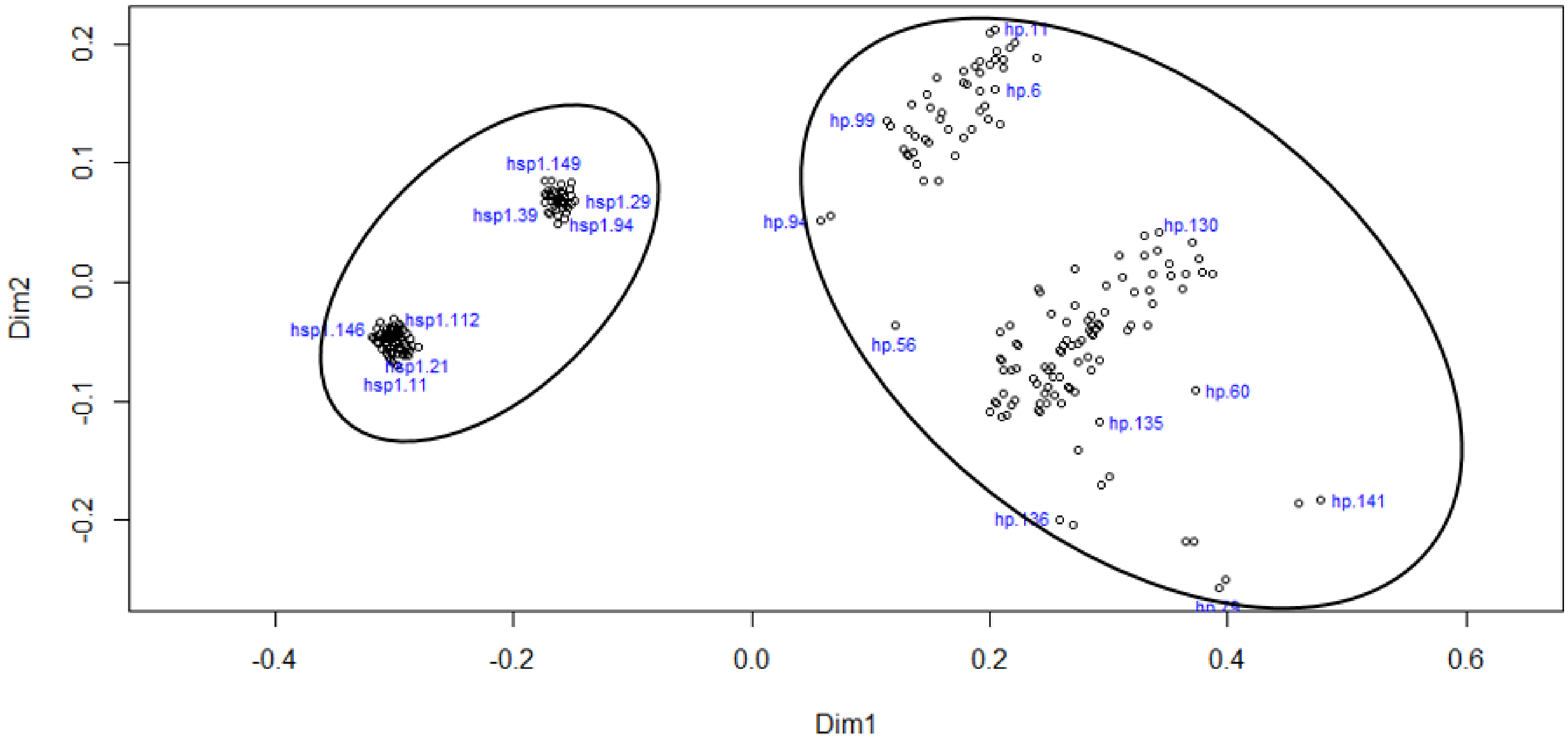
	CCA1	CCA2	CCA3	CA1
Eigenvalue	0.7926	0.1894	0.1433	0.2398
Proportion Explained	0.5806	0.1388	0.1050	0.1756
Cumulative Proportion	0.5806	0.7194	0.8244	1.0000

Figure 17. PCA ordination of NTP and BMT CVS data. The vectors drawn are for species Hydatica petiolaris and Hydatica sp1. The projections for the sites indicate a ranking of sites from low to high along each individual vector. Eigen value for Axis 1, 61.4657, Axis 2, 30.6999



	PC1	PC2	PC3	PC4	PC5	PC6
eigenvalue	61.46574	30.69986	17.79756	12.774238	10.525364	8.711511
percentage of variance	36.26459	18.11282	10.50050	7.536761	6.209932	5.139764
cumulative percentage of variance	36.26459	54.37741	64.87792	72.414679	78.624611	83.764376
broken-stick percentage	27.45343	18.36252	13.81707	10.786764	8.514036	6.695855
broken-stick cumulative %	27.45343	45.81595	59.63302	70.419782	78.933819	85.629673

Figure 18. Graph of PCoA of *Hydatica petiolaris* and *Hydatica* sp.1 petal measurements from NTP and BMT sites. Graph abbreviations: hp = *Hydatica petiolaris* individual, hsp1 = *Hydatica* sp.1 individual.



Paired t-test
t = 39.459, df = 299, p-value < 2.2e-16

Figure 19. Mesquite phylogenetic tree comparison of maximum likelihood tree and maximum parsimony trees for the 77 taxa used within this study.

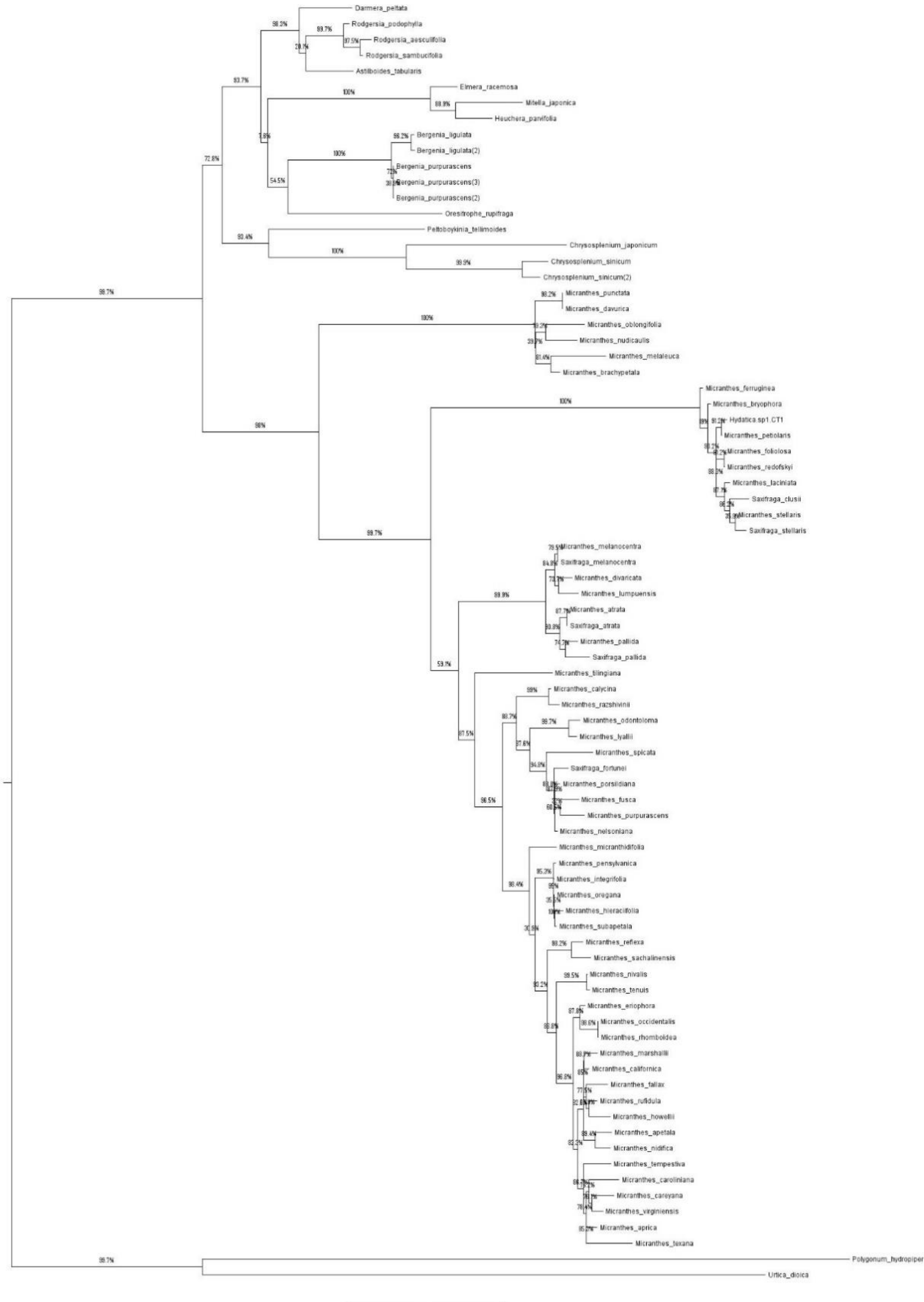


Figure 20. Phylogeny developed by Deng, et al., (2015) showing most recent phylogenetic agreement of the *Saxifragaceae* and the posterior probabilities of each group of genera. Numeric values at the nodes are Bayesian posterior probabilities obtained from the BEAST tree, with PP values P>0.70 shown. Clade names (genera/species within clades) are given on the right. Geological epoch abbreviations: Plio = Pliocene; Quat = Quaternary.

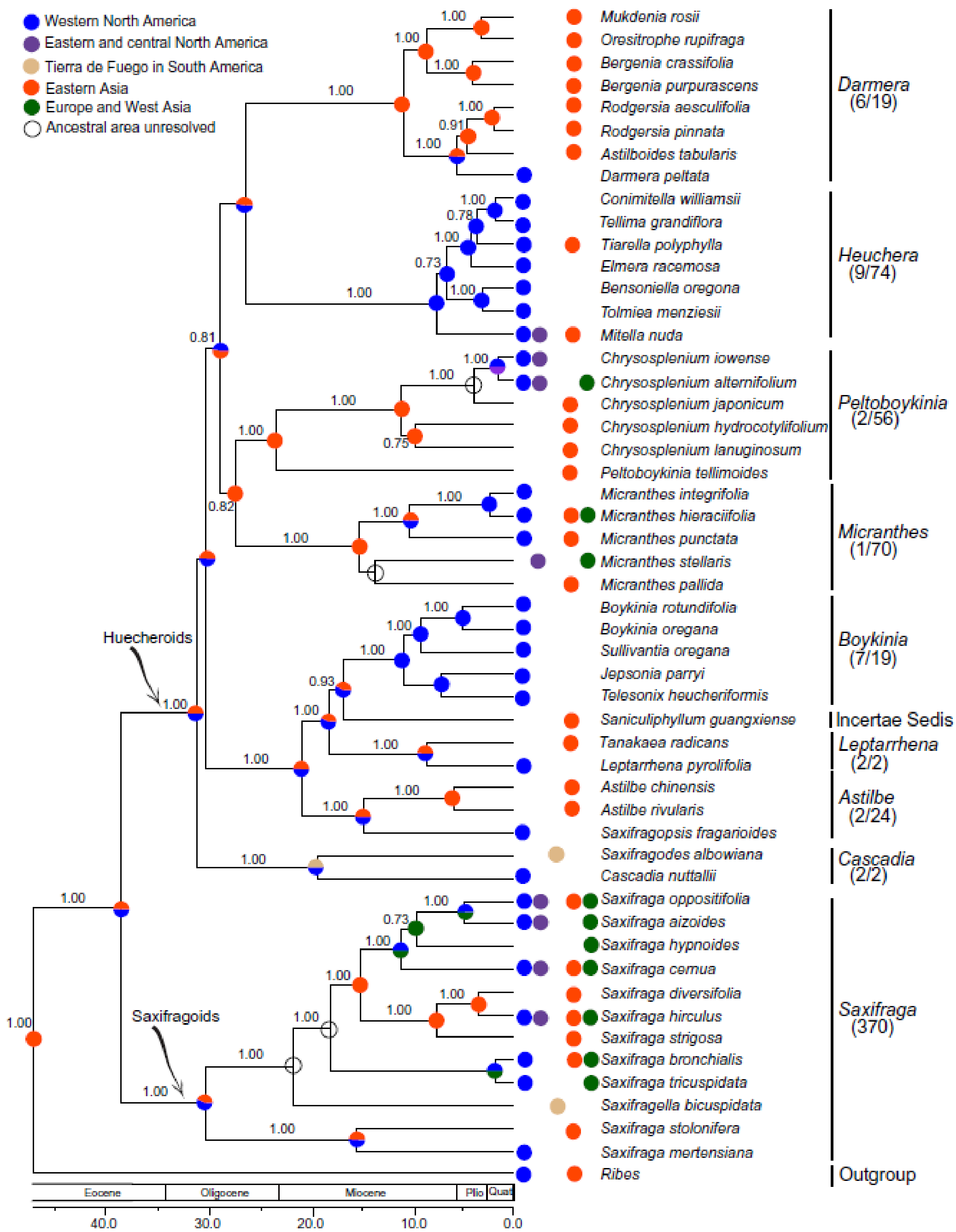


Figure 21. BLAST pairwise alignment tree of initial sequence search results with world-wide distribution. NTP sequence highlighted. BLAST computes a pairwise alignment between a query and the database sequences searched. It does not explicitly compute an alignment between the different database sequences (i.e., does not perform a multiple alignment). For purposes of this sequence tree presentation an implicit alignment between the database sequences is constructed, based upon the alignment of those (database) sequences to the query. It may often occur that two database sequences align to different parts of the query, so that they barely overlap each other or do not overlap at all. In that case it is not possible to calculate a distance between these two sequences and only the higher scoring sequence is included in the tree.

Figure 22. Maximum Likelihood Tree of initial sequence search results with world-wide distribution. NTP sequence bulleted with bootstrap score of 97.

Figure 23. BLAST pairwise alignment tree of initial sequences with local, Blue Ridge Escarpment distribution. NTP sequence highlighted. BLAST computes a pairwise alignment between a query and the database sequences searched. It does not explicitly compute an alignment between the different database sequences (i.e., does not perform a multiple alignment). For purposes of this sequence tree presentation an implicit alignment between the database sequences is constructed, based upon the alignment of those (database) sequences to the query. It may often occur that two database sequences align to different parts of the query, so that they barely overlap each other or do not overlap at all. In that case it is not possible to calculate a distance between these two sequences and only the higher scoring sequence is included in the tree.

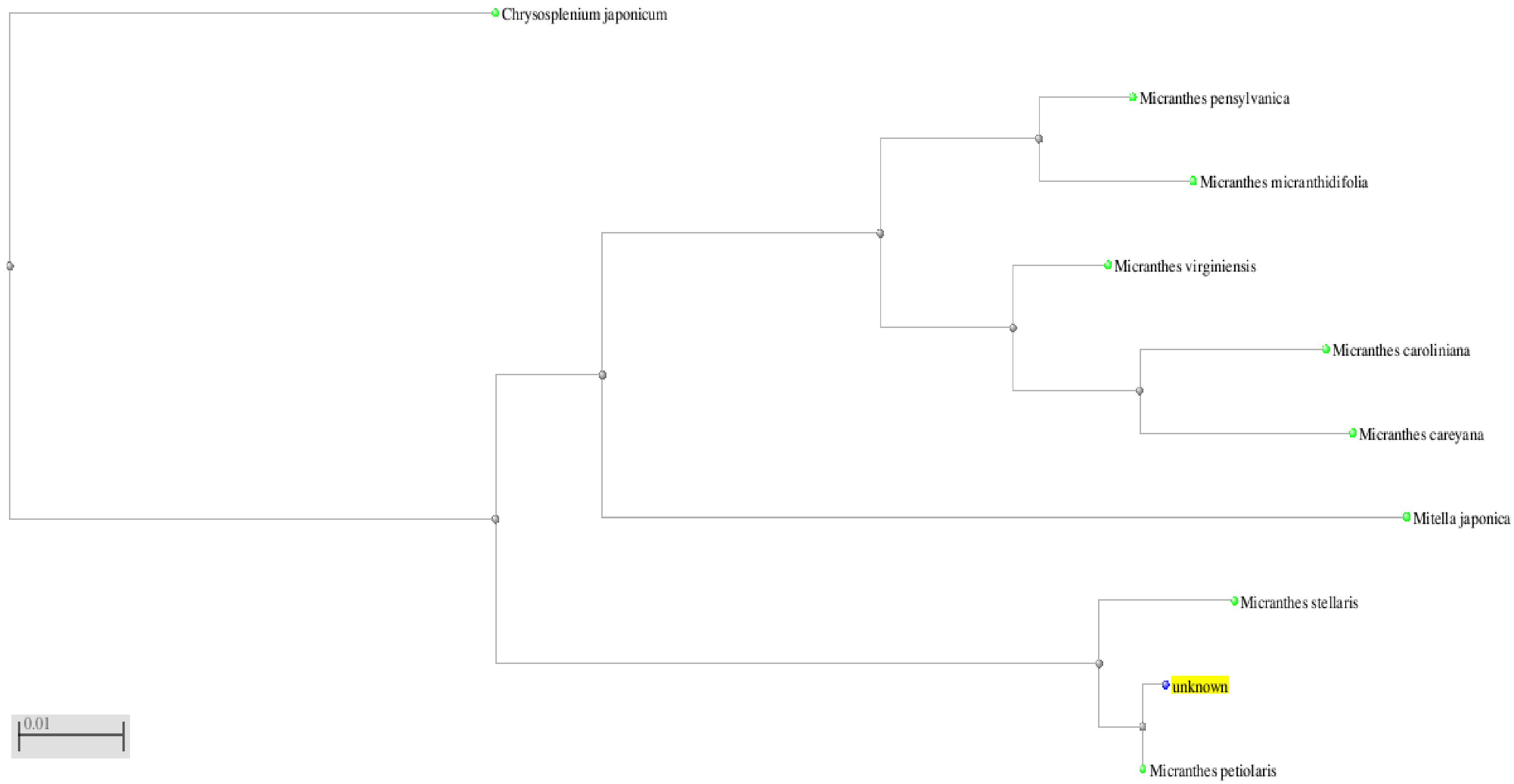


Figure 24. Reduced taxa tree representing Saxifragaceae genera found throughout the Blue Ridge Escarpment, after Weakley, et al. Numeric values at the nodes are Bayesian posterior probabilities obtained from the BEAST tree. NTP sequence bulleted with Posterior Probability score of 95.

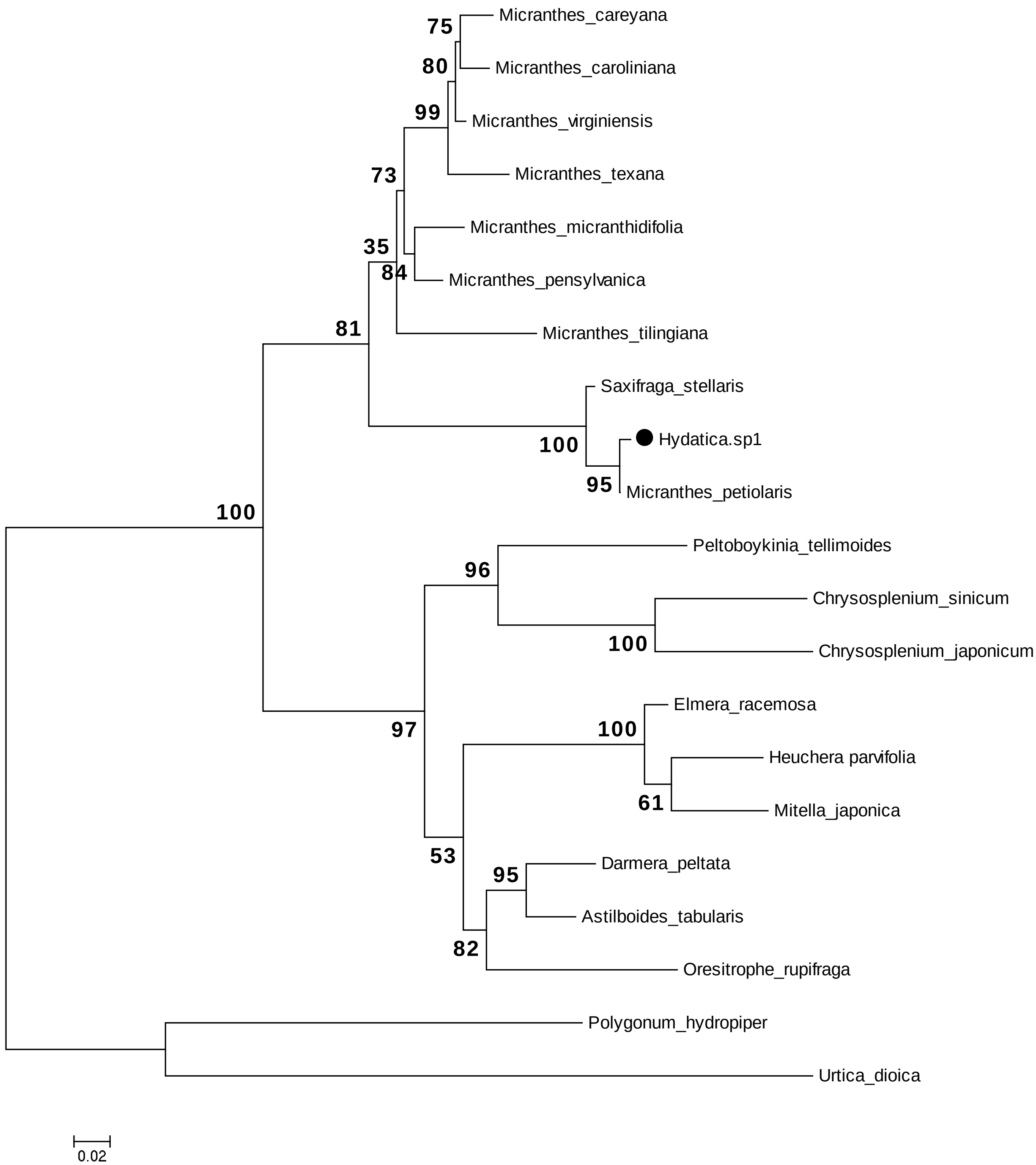
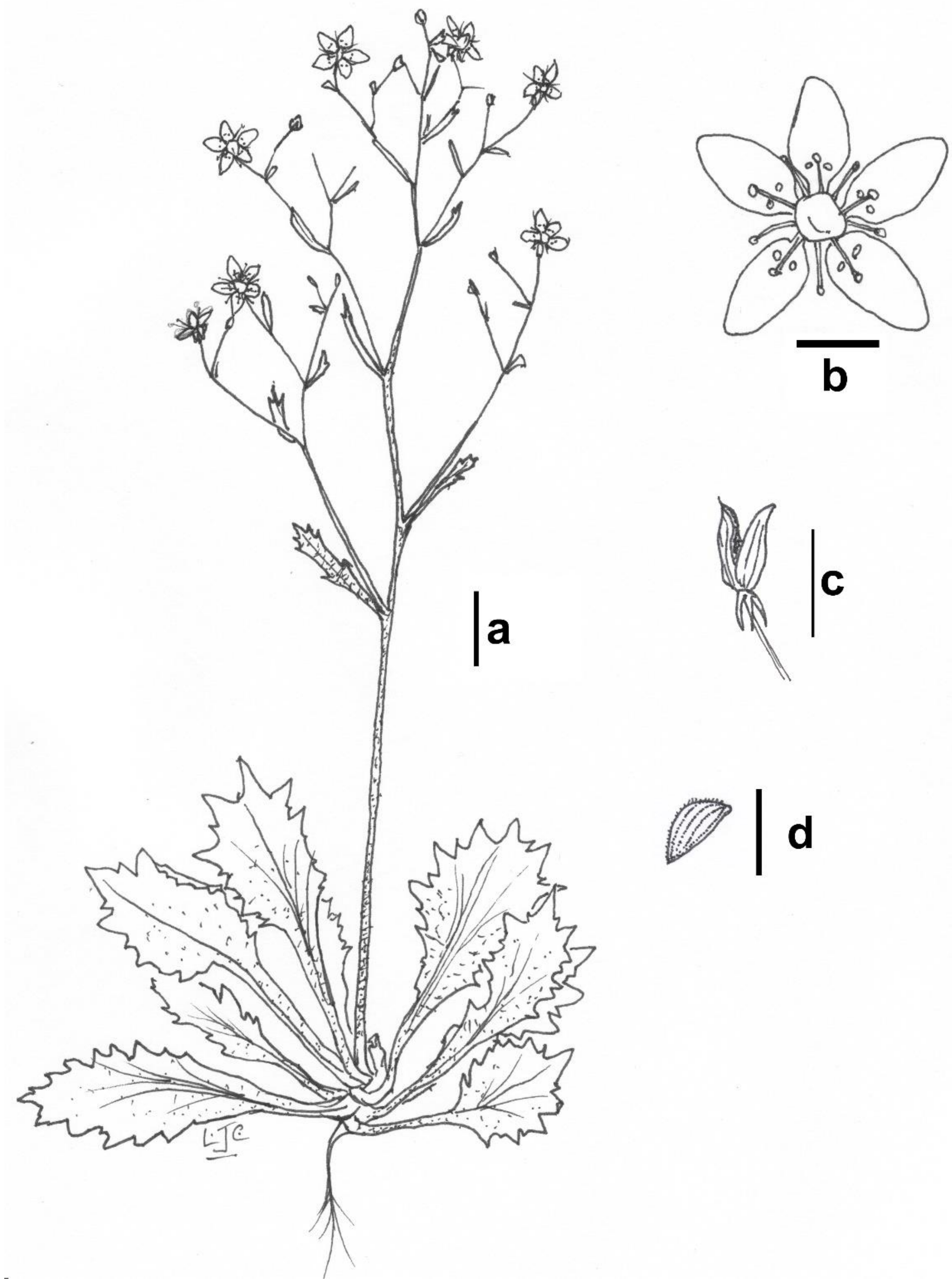


Figure 25. *Hydatica sp.1*. a: whole plant; b: flower; c: fertile, dehisced fruit; d: seed. Scale bars a = 1 cm, b, c = 5 mm, and d = 1 mm. Drawn by L. Cushman.



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APPENDICES

APPENDIX A.

DNA SEQUENCES, ITS1, 5 8S, ITS2, ALIGNED WITH MUSCLE

>Astilboides tabularis
---TCGAAA-CCTGC-A---TAGCAG-
AACAACTTGTGAACATGTGGTT-----ACACTTGGG---GAGAGGA-GTGTATGCTTT-TTCTCCC--CGCTGTCAAGA-
TGCGCTCGGTAACTTGTCAC-CCCAAAGTGC--TTGACGT---TTTG-GGGAG--GCATTATTGGGTG--CT-C---TCGACTT-AACAAC-
GAACCCCGGCGTGAATTGCGCCAAGGAA--TTT---AAAGAA-----AGAGCA--CTCCTCCATTGTTTTGTGTTCGT----GCAGACAA-
TTGGAGGAAGTGT--CGTCTTCTTGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTTTTTGAACGCAAGTTGCGCCCGAAGCCATTTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACACACTTCTCCCCAC
-AAACCTTTCCCTC---TTCGGG-TCAATGAGT-TTGTGTCGGGAAGATAC-------TGGCCTCCCG-------TT-
CACAACATTGGTGCGGTTGACCTAAAAAAGAGTACCGA-GTGGCAAAATGTCACGATAAGTGGTGGTTTATAATCC--TAATGTGG-
TTTTGCC----AAATCGAGTCGTGAG-CGTTTGTCAC-TCGAGAT--TAGCTCAA------------------GTG-AACCCCC-ACACG--
TCTAACTGATGCTATTGTCGCGACCCCA--
>Bergenia ligulata
---GGATCATTGTCGAA--CCTGC-A---TAGCAG-
AACAACTTGTGAACACGTAGTT---ACAATTTGGGG-G-AGGAGGA-GCGCATGCTTG-TTCTCCC--CGTTGTCAAGA-
CGTACTCGGTAACCTGTCGC-CCTTTAGCGC--TTTGCGT---TAGG-GGAGG--CATTATTTGGGTG--CT-C---TTGACTT-AACAAC-
GAACCCCGGCGTGAATTGCGCCAAGGAATTT-----AAAGAA-----AGAGCA--TTCCTCCATTG------CTGCTCTAT-GCGGACGA-
TTGGAGGAGGTGT--TATCTTCTTGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTCTTTGAACGCAAGTTGCGCCCGAAGCCATTTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTTAACACATCT-
CCCAC-AAACCCTTCCCCATGTCTTGGG-GCAAGGATT-TTGTGGCGAGAAGA-AT-------TGGCCTCCCG-------TG--
TAAACATTGGCGCGGTTGACCTAAAAAAGAGTGCCGA-GTGATGAAATGTCACGATAAGTGGTGGTTTTTAATCC--TAATGTGA-
TTTTGCC----GAATCGAGTCGTGAT-CATTTGTCAC-TGCAGAT--ATGCTCAA------------------GTG-AACCCCA--TACG--
TCTAGCTGACGCTATG-TCG--
>Bergenia purpurascens
--TTTCCGTAGGTGAACCTGCGGAAGGATCATTGTCGAAA-
CCTGC-A---TAGCAG-AACAACTTGTGAACATGTAGTT---ACAATTTGGGG---AGGAGGA-GCGCATGCTTG-TTCTCCC--CGTTGTCAAGA-
CGTACTCGGTAACATGCCGC-CCTTTAGTGC--TTTGCGT---TAGG-GGAGG--CATTATTTGGGTG--CT-C---TTGACTT-AACAAC-
GAACCCCGGCGTGAATTGCGCCAAGGAATTT-----AAAGAA-----AGAGCA--TTCCTCCATTG------CTGCTCTAT-GCGGACAA-
TTGGAGGAGGTGT--TATCTTCTTGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTCTTTGAACGCAAGTTGCGCCCGAAGCCATTTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTTAACACATCT-

CCCAC-AAACCCTTCCCCATGTCTTGGG-GCAAGGATT-TTGTGGCGAGAAGA-AT-------TGGCCTCCCG-------TGTAAC---
ATTGGCGCGGTTGACCTAAAAAAGAGTGCCGA-GTGATGAAATGTCACGATAAGTGGTGGTTTATAATCC--TAATGTGA-TTTTGCC----
GAATCGAGTCGTGAT-CATTTGTCAC-TTCAGAT--ATGCTCAA------------------GTG-AACCCCA--TACG--
TCTAGCTGACGCTATTGTCGCGACCCCA-GTCAGGCGGGAATACCCGCTGAGTTTAA---
>Chrysosplenium japonicum
---TCCGGAGGTGAACCTGCGGAAGGATCATTGTTGTAA-
CCTGC-C---TAGCAG-CGCAACTCGTGAACACGTAACT-----AAATGTGGGGGGAGAAGTG--TGCAAGCTCT-TTCTTTCCTCGTTTCAAGTA-
-GTACCCGGTATATC------CCACACACGG-CTTAATGC--CTTGG-TGGAG--TAATTGCCGGATG--CT-----CTTGTGCTAACAA--
AACCCACGGCGTGAATTGCGCCAAGGAA-TTTTATGAA---------AGATCA--TTCTTCCATTG--GTGTGTTC---
ATCGCATCAAAATTGGAGGAGTTGT--CATCTTCGTGATGTCTC-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTTTTTGAACGCAAGTTGCGCCCAAAGCCATTTGGTTGAGAGCACGTCTGCCTGGGCGTCACGTACACACATCTCGCCA
C-AAACCATTACCAAGCTTATTGG-TGCATGAGT-TGGTGGTGAGCCAATAT-------TGGTCTCCCG-------TGCTGC---
AATGTTGCGGTTGACCTAAAAAAGAGTACCTA-GTGACCGGACGTCACGATAAGTGGTGGTT--TAATCC--AATTATGG-TTTTGCC----
GAATCGAGTCGTGAA-TGTTTGTCACTTTGGAAA--GTGCTAGA------------------GTTTGAACCCC-ACACA-
TCTCAATGGATGCTATTGTCGCGACCCCAGGTCAGGCGGGATTACCCGCTGAGTTTAAGCATATCAATAAGCGGAGG---------------------------
----
>Chrysosplenium sinicum
--TCGTAA-CCTGC-C---TAGCAG-
TATGACTCGTGAACATGTGACCAATACACGTGGGGGGGCAGAAGTG--CGCAAGCTCT-TTTTCCC--CTCTTCGGGCA--
GCACTCGGTGTTTCCCCAC----ACATTGC--TTTATGC--CTTGG-TGGAG--TCATCACCGGATG--CT-C---TCGGCCT-AACCA--
AAACCACGGCGTGAATTGCGCCAAGGAA-TTTTATGAA---------AGATCA--TTTCTCCGTCG--GTGTGTTTTC----ACATTCGA-
TAGGAAGATGTGT--CATCTTCGTGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTTTTTGAACGCAAGTTGCGCCCGAAGCCATTTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACACACATCTCGCCA
CAAACGCTTCACTCA---CTGTTGGTGCGTGAGT-TTGTGACGAGACGATAC-------TGGTCTCCCG-------TGATGCAACGT---
TGCGGTTGACCCAAAAATGAGTACCTG-GTGACGAGACGTCACGATAAGTGGTGGTT--TAAGCC--AATTTTGG-TTTTGCC----
GAATCGAGTCGTGAGTTGTTTTTCGCTTTGAAAT--GTACTCGA------------------GAT-GAACCCC-ACACG--TCT--
----
>Darmera peltata
--TCGAAA-CCTGC-A---TAGCAG-
AACAACTTGTGAACATGTGGTT-----ACAATTGGG---GAGAGGA-GTGCATGCTTT-TTCTCCC--CACTGTCAAGA-
TGTGCTCGGTAACTTGTCGG-CCCAAAGTGC--TTGACAT---TTTG-GGGAG--GCATTATTGGGAG--CT-C---TTGACTT-AACAAT-
GAACCCCGGCGTGAACTGCGCCAAGGAA--TTT---AAAGAA-----AGAGCG--CTCCTCCATTGTTTTGTGTTCGT----GCAGACTA-
TTGGAGAAAGCGT--CGTCTTCTTGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTCTTTGAACGCAAGTTGCGCCCGAAGCCATTTGGTCGAGGGCACGTCTGCCTGGGCGTCACGTACACACATCT-

CCCAC-AATCCTTTCCCCT---TTTTGGGTGAAGGAGT-TTGTGTCGGGAAGATAT-------TGGCCTCCCG-------
TGCACAAATATTGGTGCGGTTGTCCTAAAAAAGAGTACCGA-GTGATGAAATGTCACGATAAGTGGTGGTTTATAATCC--TAATGTGG--
TTTGCC----AAATCGAGTCGTGAG-CGTTTGTCAC-TCGAGCT--AAGCTCAA------------------GTTAAACCCCC--CACG--TCT------------------------
----

>Elmera racemosa

--- -------TCGAAA-CCTGC-A---TAGCAG-
AACAACTTGTGAACATGTAGTT---AC--ACTTGGGAGGGGAGGGA-GTGCATGCTCT-CTCTCCCC-CGTTGTCAATA-
TGTGCTCAGTAACCTGCCGC-CCGAAAGCGC--TTGGCGC---TTGA-AGGAG--GCGTTATTGGATG--CT-C---TTGACTC-AACAAC-
GAACCCCGGCGTGAATTGCGCCAAGGAA----TACAAAAGAA-----TGACCA--GTCCTCCATTT-GCTGTGTTCTTTGC-ACAAGTGA--
TTGGAGTATTGT--CATCTTCTTGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
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CCCACAAAACCTTTCCCCA---TTTTGG-GATCAGGATTTTGTGGCGAGCAGAGAG-------TGGCCTCCCA-------TG---
CAGTTTGTGTGTGGTTGACCCAAAAAAGAGTACCGA-GTGAYGAAGTGTCACGATAAGTGGTGGTTTCTAAGCC--CACAGTGG-
CTTTGCC----AAATCGAGTCGTGTG-CATTTGTTGC-TCGGGAT-GATTCTCAA------------------GCG-AACCCCA-ACACG--TCT---------------------
----

>Heuchera parvifolia

--- -------TCGAAA-CCTGC-A---TGGCAG-
AACAACTTGTGAACATGTAGTT-----AAACTTGGG-GGGGAGGGA-GTGCACGCTCG-CTCTCCCCCCGTTGTCAATG-
TGTGCTCAGTAACCTCCCGC-CCGAAAGCGC--CTGGCGC--TTGAA-GGAG---GCGTTATTGGATG--CTCT---TTGACTT-AACAAC-
GAACCCCGGCGTGAATTGCGCCAAGGAA----TACAAAAGAA-----TGATCA--GTCCTCCATTT-GCTGTGTTCTT----
GCACAAGCGACTGGGGTATTGT--CATCTTCTTGATGTCTT-
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GAACCATCGAGTTTTTGAACGCAAGTTGCGCCCGAAGCCATCTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACACACGCCT-
CCCACAAAACCTTTCTCCA---TTTTGG-GACAAGGATTTTGTGGCGAGCAGAGAG-------TGGCCTCCCG-------TG---
CAAMTTTTGTGCGGTTGACCCAAAAAAGAGTACCGA-GTGACGAAGTGTCACGATAAGTGGTGGTTTGTAAGCC--CTTAGTGG-
CTTTGCC----AAATCGAGTCGTGTG-CATTTGTTGC-TCGGGAT-GATCCTCAA------------------ACG-AACCCCT-ACACG--TCT----------------------
----

>Hydatica sp1

TTTTAGGAGAAGTCGTAACAAAAGTTTCCGTAAGGTGAAACTGCGGAAGGATCATTGCTGAAA--CTGC-A---ATGCAG-
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GTC-GTGGGCTTGC-TTCCATGC--ATGCA-CGTCG--GAGT-ATTGAGCG--TT-C---CCTGCAA-TGTAAC-
AACCCCCGGCGCGAATTGCGCCAAGGAATTTTTAAAAAAGAG-----AGGGCA-CTTTCTCCACTT---CATGCC----AT-GCACAAAG--
TGGAGAGAATGT--TATCTTCTAGATGTCTT-
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GAACCATCGAGTTTTTGAACGCAAGTTGCGCCTGAAGCCTCTTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACACACTTCT-CCCAC-

AATCC-TTTCACA----TGAAG-TGTTTGCGT-TTGCGGGGAGGGATGAT-------TGGCCTCCCA-------TG-CCC---
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TCGCGACCACAGGTCAGGCGGGATTACCCGCTGAG-TTAAGCTTCCCATATAGGCGGGAGGAAAAAAAACAACG------------------
>Micranthes apetala
--AAA-CCTGC-A---ATGCAG-
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>Micranthes aprica
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>Micranthes atrata
--GATGAAA-CCTGCCA---AAGCAG-
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>Micranthes brachypetala

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TTG-GGGAA--GAAT-GTTGAGTA--CC-C---CCGACTT-CAAAAC-TCACCCCGGCGTGAATTACGCCAAGGAC-TTTTTAAAAGGAT-----
TGAACA--TTCCTTCACATTACTGTATTCAT----GCAGGTAA-TTGGAGGAGGTGT--CATCTTCTTGATGTCTT-
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TTGCGACCCC---
>Micranthes bryophora
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>Micranthes californica
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TCGCGACCCCAGGTCAAGCGGGATTACCCGCTGAGTTTAA--
>Micranthes calycina

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TCGCGACCCCA---
>Micranthes careyana

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>Micranthes caroliniana
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TCGCGACCCC--
>Micranthes davurica

---ACC-CCATG------CTTTATGC--TTGGG-GAAGA--AAAT-GTTGATTA--CC-C---
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GCATGTAA-TTGGAGGAGGTGT--TATCTTCTTGATGTCTT-
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TAATCGAGTCGAG---CGTTTATCAC----AAAT--GTGCTCCC------------------A---AAACCCAAACACG--TTGATTTGGCGCTATT-
TTGCGACCCC--
>Micranthes divaricata
---TTGATGCA--CCTGCTA---AAGCAG-
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TGCAACAAACCCCCGGCGTGAATTGCGCCAAGGAA---TTTTAAAAAAAGATAGATAACA----CCTCCATCT---CATGTT-------------------
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>Micranthes eriophora
---ATTGCTGAAACCCTGC-A---ATGCAG-
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>Micranthes fallax
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>Micranthes ferruginea
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>Micranthes foliolosa

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GCGACCCCA---

>Micranthes fusca
-- ----TTGCTGAAA-CCTGC-A---ATGCAG-AAAACCTTGAGAACATGTAA-----AAAATATTGAG-G-AGGGGCA-TTTCATGTTTC-TTCTCAC--CTTTGTTGGGT-GTCATTCGATTTTTTTAACC-CTGCATTTGC-TTTTACGCATGTGTA-GGTGC--AAATAATTGAATG--TT-C---CTTACAT-TGCAAC-AACCCCCGGCGTGAATTACGCCAAGGAA---TTTTAAAAAAA-----AGAGCA--TTTCTTCATTT---CATGTGTAT----GCATTAA---TTGGAAAAAATGT--AATCTTATTTATGTCTT-TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGTGAACCATCGAGTTTTTGAACGCAAGTTGCGCCCGAAGCC-TCTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACACACTTCT-CCCAC-AAACC-TTTCCCA---TTTTGG-TTTAGGCAT-TTGTGATGAGGAAAGAT-------TGGCCTCCCA-------TA-CCCTCCTTGCGTGTGGTTGACCTAAAAAAGAGTA-CGA-GTGATGAAACGTCACGATAAGTGGTGGTTTATAAGCC--TTTTTTGG-TTTTGCC----AAATCGAGTCGAA---CGTTTGTCAC-TTGAAAT--ATGCTCAA------------------GTT-AATCCCC-ACACG---TGAATTGACGCTATT-TCGCGACCCCA--
>Micranthes hieraciifolia
-- ----------------TGC-A----------GAAACCTTGAG-ACATGTTA-----AAAAAATTGAG-G-AGGGGCA-TTTCATGTTTC-TTCTCAC--CTTTGTGGGGA-GACATTTGATTTCTTTAACC-TTGYGTTTGC-TTTTACGC--ACGCG-GGTGT--AAGTAATTGAGTG--TT-C---CTTACAT-TKTAAC-AACCCCCGGCGTGAATTGCGCCAAGGAATTTTT---AAAAAA-----AGAGCA--TTTTTTCATTT---TATGTTAAT-AT-GCATAGA---TTGGGAAAATGT--TATCTTCTTTATGTCTT-TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGTGAACCATCGAGTTTTTGAACGCAAGTTGCGCCCGAAGCC-TYTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACACACTTCT-CCCAC-AAACCTTTTCCCA---TTTTGG-TTTAAGAAT-TTGTAGTGAGAAAAGAT-------TGGCCTCCCA-------TACCCC---TTGTGTGTGGTTGACCTAAAAAAGAGTA-CGA-GTGATGAAACGTCACGATAAGTGGTGGTTTATAARCC--TTTTTTGG-TTTTGCC----AAATCGAGTCGAA---CGTTTGTCAC-TTAAGAT--ATGCTCGA------------------GTT-AATCCCC-ACACG--TTGAATTGACGCTATT-TCGCGACCCCA--
>Micranthes howellii
-- --CATTGCTGAAA-CCTGC-A---ATGCAG-AAAACCTTGAGAACATGTTA-----AAAAAATTGAG-G-AGAGACA-TTTCATGTCTC-TTCTCGC--CTTTGTGGGGA-GGCATTTGATTTCTTTAACC-TTGCGTTTGCTTTTTATGC--ATGCA-TGTGT--GAGTAATTGAGTG--TT-C---CTTACAT-TGTAAC-AACCCCCGGCGTGAATTGCGCCAAGGAA--TTTTAAAAAAAAA-----AGAGCA---TTTTCCATTT---TATGTTATT-AT-GCATAAAA--TGGA-AAAATGT--TATCTTCTTTATGTCTT-TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGTGAACCATCGAGTTTTTGAACGCAAGTTGCGCCCGAAGCC-TCTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACACACTTCT-CCCAC-AAACC-TTTCCTA---TTTAGG-TTTAAG-AT-TTGTGATGAGAAAAGAT-------TGGCCTCCCA-------TA-CCC---TTACGTGTGGTTGACCTAAAAAAGAGTA-CGA-GTGATGAAACGTCACGATAAGTGGTGGTTCATAAGCC--TTTTTTGG-TTTTGCC----AAATCGAGTCGCA---CGTTTATCAC-TTGACAT--ATGCTCGA------------------GTT-AATCCCC-ACACG--TTGAAATGACGCTATT-TCGCGACCCCAGGTCAGGCGGGATTACCCGCTGAGTTTAAGCATAT---
>Micranthes integrifolia

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>Micranthes laciniata
--AA-CCTGC-A---ATGCAG-
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AATCC-TTTCACA----TGTAG-TGTTTGCGT-TTGCGGCGAGGGATGAT-------TGGCCTCCCA-------TG-CCT---
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TCGCGACCCCAGGTCAGGCGGGATTACCCGCTGAGTTTAAGCATATCA---
>Micranthes lumpuensis

GAGAACATGTAAC----AAGAAATTGAG-T-GGGAGCA-TTCCATGCTTC-TTCTCAC--CTTTGTCGGGA-GGCATTAGATCCATTGCTCC--
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TGCAACAAACCCCCGGCGTGAATTGCGCCAAGGAA---TTTTAAAAAAAGGTAGATAGCA----CCTCCATCT---CATGTT------------------
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TCGCGACCCCAGGTCAGGCGGGACTACCCGCTGAGTTTAAGCATATCA---
>Micranthes lyallii

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TTGCGACCCC--
>Micranthes marshallii
---ATTGCTGAAA-CCTGC-A---ATGCAG-
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AAATCGAGTCGAA---CGTTTATCAC-TTGAGAT--ATGCTCGA-----------------GTT-AATCCCC-ACACG--TTGAAATGACGCTATT-
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>Micranthes melaleuca

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TTGGG-GAAGA--AAAT-GTTGTGTA--CG-C---CCGACTT-CAAAAC-TCACCCCGGCGTGAATTACGCCAAGGACTTTTT-AAAAGGAT-----
TGAACA--TTCCTTCACATTACTGTATTC---AT-GCAGATAA-TTGGAGGAGGTGT--TATCTTCTTGATGTCTT-
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TTGCGACCCC--
>Micranthes melanocentra

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TCGCGACCCCAGGTCAGGCGGGATTACCCGCTGAGTTTAAGCATATCA--
>Micranthes micranthidifolia
--- -----TGCTGAAA-CCTGC-A---ATGCAG-
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TCGCGACCCCAGGTCAGGCGGGATTACCCGCTGAGTTTAAGCATATCA--
>Micranthes nelsoniana

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>Micranthes nidifica

---TTGCTGAAA-CCTGC-A---ATGCAG-
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>Micranthes nivalis
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>Micranthes nudicaulis

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>Micranthes oblongifolia

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>Micranthes occidentalis

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>Micranthes odontoloma

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>Micranthes oregana

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>Micranthes pallida
--TTGATGAA--CCTGCCA---AAGCAG-
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>Micranthes pensylvanica
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>Micranthes petiolaris

--GAAA-CCTGC-A---ATGCAG-
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GTC-GTGGGCTTGC-TTCCATGC--ATGCA-CGTCG--GAGT-ATTGAGCG--TT-C---CCTGCAA-TGTAAC-
AACCCCCGGCGCGAATTGCGCCAAGGAATTTTTAAAAAAGAG-----AGGGCA-CTTTCTCCACTT---CATGCC----AT-GCACAAAG--
TGGAGAGAATGT--TATCTTCTAGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTTTTTGAACGCAAGTTGCGCCTGAAGCCTCTTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACACACTTCT-CCCAC-
AATCC-TTTCACA----TGAAG-TGTTTGCGT-TTGCGGGGAGGGATGAT-------TGGCCTCCCA-------TG-CCC---
ACGTGTGTGGTTGGCCTAAAAATGAGTA-CTA-GTGATGAAACGTCACGATAAGTGGTGGTACATAATCC--TTCTTTGGTTTTTGCC----
AAATCGAGTCGTA---AGTTTGTCGC-TTGAGAA--ATACTCAA-----------------GTT-AATCCCC-TTACG--TTGATCATACGTTATT-
TCGCGACCCCAGGTCAGGCGGGATTACCCGCTGAGTTTAAGCATATCA---
>Micranthes porsildiana

ACAAGGTTTCCGTAGGTGAACCTGCGGAAGGATCATTGCTGAAT-CCTGC-A---ATGCAG--AAAACTTGAGAACATGTAAAA---
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TTTTACGCATGTGTG-GGTGT--AAGTAATTGAGTG--TT-C---CTTACAT-TGCAAC-AACCCCCGGCGTGAATTGCGCCAAGGAATTTTT-
AAAAAAAG-----AGAGCA--TTTCTCCATTT---CATGTGTAT----GCATTAA---TTGGAAAAATGT--AATCTTATTTATGTCTT-
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GAACCATCGAGTTTTTGAACGCAAGTTGCGCCCGAAGCC--TCGGTTRAGGGCACGTCTGCCTGGGCGTCACGTACACACTTCT-CCCAC-
AAACC-TTTCCCR---TTTTGG-TTTAGGCAT-TTGTGATGAGGAAAGAT-------TGGCCTCCCA-------TA-
CCCTCCTTGCGTGTGGTTGACCTAAAAAAGAGTA-CGA-GTGATGAAACGTCACGATAAGTGGTGGTTTATAAGCC--TTTTTTGG-
TTTTGCC----AAATCGAGTCGAA---CGTTTGTCAC-TTGAAAT--ATGCTCAA-----------------GTT-AATCCCC-ACACG---
TGAATTGACGCTATT-TCGCGACCCC---
>Micranthes punctata

ACAAGGTTTCCGTAGGTGAACCTGCGGAAGGATCATTGCCGAAA-CCTGC-A---TAGCAG-AAAACCAAGTGAACTTGTAGA----
AAAACTCTGAG---GGAAGCA-TCACGTGCTTC-TTCTCCA--TGTTGTCGGGA-TGTGCTTGATAGTTTCCACC-CCATG------CTTTATGC---
TTGG-GGAAG--AAAATGTTGATTA--CC-C---CCGACTT-CAAAAC-TCACCCCGGCGTGAATTACGCCAAGGACTTTTT-AAAAGGAT-----
TGAACA--TTCCTCCACATTACTGTATTCAT----GCATGTAA-TTGGAGGAGGTGT--TATCTTCTTGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCACAACGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTTTTTGAACGCAAGTTGCGCCCGAAGCCATTTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACCCACTCCT-CTCAC-
AACCTATATTCCA-----TCGGTCTTAGGGHT-TTGTKTTGAGGAAAGGT-------TGGYCTCCCA-------TGCACT---
ATGTGTGTGGTTGTCCTAAAATAGAGTACCGA-GTGATAAAACGTCACGATAAGTGGTGGTATACAAA----------------GCC----
TAATCGAGTCGAG---CGTTTATCAC----AAAT--GTGCTCCC-----------------A---AAACCCAAACACG--TTGATTTGGCGCTATT-TTGCG--------
----
>Micranthes purpurascens

---CATTGCTGAAA-CCTGC-A---ATGCAG-
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GTCATTTGATTTTTTAACCC--TGCATTTGC-TTTTACGCATGTGTA-GGTGT--AAGTAATTGAGTG--TT-C---CTTACAT-TGCAAC-
AACCCCCGGCGTGAATTGCGCCAAGGAA-TTTTTAAAAAAAG-----AGAGCA--TTTCTCCATTT---CATGTGTAT----GCATTAA---
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TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTTTTTGAACGCAAGTTGCGCCCGAAGCC-TCTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACACACTTCT-CCCAC-
AAACC-TTTCCCA---TTTTGG-TTTAGGCAT-TTGTGATGAGGAAAGAT-------TGGCCTCCCA-------TA-
CCCTCATTGTGTGTGGTTGACCTAAAAAAGAGTA-CGA-GTGATGAAACGTCACGATAAGTGGTGGTTTATAAGCC--
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>Micranthes razshivinii
---AAA-CCTGC-A---ATGCAG-
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AACCCCCGGCGTGAATTGCGCCAAGGAATTTTT-AAATTAAG-----AGAGCA--TTTCTCTATTT---CATGTTTAT----GCATAAAC--TGGA-
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AAACT-TTTCCCA---TCTTGG-TTTAGGCAT-TTGTGTTGAGAAAAGAT-------TGGCCTCCCA-------TA-CCC---
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TCGCGACCCCAGGTCAGGCGGGATTACCCGCTGAGTTTAAGCATATC---
>Micranthes redofskyi

ACAAGGTTTCCGTAGGTGAACCTGCGGAAGGATCATTGCTGAAA-CCTGC-A---ATGCAG-AAAACCGTGTGAACGCGTGACC---T-
AAAAAAGAG-A-AGGAGCA-TTCTTTGTTCC-TTTTTAC--CTTTGTTGGAT--TCGCTTCATTCCTTT-GTC-GTGGACTTGC-TTCCATGC--
ATGCA-CGTCG--GAGT-ATTGAGCG--TT-C---CCTGCAA-TGTAAC-AACCCCCTGCGCGAATTGCGCCAAGGAAATTTTAAAAAAGAG-----
AGGGCA-CTTTCTCCACTT---CATGCC----AT-GCACAAAG--TGGAGAGAATGT--TATCTTCTAGATGTCTT-
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AATCC-TTTCACA----TGAAG-TGTTTGCGT-TTGCGGGGAGGGATGAT-------TGGCCTCCCA-------TG-CCC---
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AAATCGAGTCGTA---AGTTTGTCGC-TTGAGAA--ATACTCAA------------------GTT-AATCCCC-TTACG--TTGATCATACGCTATT-T-------------
----
>Micranthes reflexa

ACAAGGTTTCCGTAGGTGAACCTGCGGAAGGATCATTGCTGAAT-CCTGC-A---ATGCAG-AAAACTTTGAGAACATGTTA-----
AAAAAATTGAG-G-AGGGGCA-TTTCATGTTTC-TTCTCAC--CTTTGCTGGGA-GGCATTTGATTTCTTTAACC-TTGCGTTTGC-TTTTTTGC--
ATGCG-GGTGT--AAGTAATTGAGTG--TT-C---CTTGCAT-TGCAAC-AACCCCCGGCGTGAATTGCGCCAAGGAA--TTTTTAAAAAAA-----
AGAGCA-TTTTTTTCATTT---TATGTTAAT-AT-GCATAAAA--TAGA-AAAATGT--TATCTTCTTTATGTCTT-
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AAACC-TTTCCTA---TTTCGG-TTTAAGAAT-TTGTGGTGAGGAAAGAT-------TGGCCTCCCA-------TA-CCC---
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TCGCGACCCCAG---
>Micranthes rhomboidea
--CTGAAACCCTGC-A---ATGCAG-
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TAAATGT--TATCTTCTT-ATGTCTT-
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>Micranthes rufidula
---TTGCTGAAA-CCTGC-A---ATGCAG-
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>Micranthes sachalinensis

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ATGCG-GGTGT--AAGTAATTGAGTG--TT-C---CTTGCAT-TGCAAC-AACCCCCGGCGTGAATTGCGCCAAGGAATTTTT---AAAAAA-----
GGAGCA--TTTTTTCATTT---TATGTTAAT-AT-GCATAAAA--TTGA-AAAATGT---ATCTTCTATATGTCTT-
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TCGCGACCCCA---
>Micranthes spicata
---AAGGTTTCCGTAGGTGAACCTGCGGAAGGATCATTGCTGAAA-
CCTGC-A---ATGCAG-AAAACCTTGAGAACCTGTAAA----AAAATATTGAG-A-AGGGGCA-TTTCATGCTTC-TTCTCAC--CTTTGTTGGGA-
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TTTTGCC----AAATTGAGTCGAA---CGTTTTTCAC-TTGAAAT--ATGCTCAA------------------GTT-AATCCCC-ACACG---
TGAATTGACGCTATT-TCGCGACCCCA---
>Micranthes stellaris

ACAAGGTTTCCGTAGGTGAACCTGCGGAAGGATCATTGCTGAAA-CCTGC-A---ATGCAG-AAAACCGTGTGAACACGTGACC---
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ATGCA-CGTCG--GAGT-ATTGAGTG--TT-C---CCTACAA-TGTAAC-AACCCCCGGCGCGAATTGCGCCAAGGAATTTTTAAAAAAGAG-----
AGGGCA-CTTTCTCCACTT---CATGCC----AT-GCACAAAG--TGGAGAGAATGT--TATCTTCTAGATGTCTT-
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AAATCGAGTCGTA---AGTTTGTCGC-TTGAGAA--ATACTCAA------------------GTT-AATCCCC-TTACG--TTGATCATACGCTATT-
TCGCGACCCCA---
>Micranthes subapetala

---GAACCCTGC-A---ATGCAG-
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----
>Micranthes tempestiva
--TTGCTGAAT-CCTGC-A---ATGCAG-
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>Micranthes tenuis

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TCGCGACCCCA---
>Micranthes texana

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>Micranthes tilingiana
---GC-A---ATGCAG-
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>Micranthes virginiensis
---TTGCTGAAACCCTGC-A---ATGCAG-
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>Mitella japonica

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>Oresitrophe rupifraga
---GCAA---CAGCAG-
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CCCAC-AAACCTTTTCCCA---TATTGGGTAAAATGATTTGGCGGCGAGTTGAGAT-------TGGCCTCCCG-------TG--
CAAACAATTGTGCGGTTGACCTAAAACAGAGTACCGA-GTGATGAAATGTCACGATAAGTGGTGGTTTATAATCA--TAATGTGG--
TTTGCC-----AATCGTATCGCGAT-CATTTGTTAC-TCGAGGC--ACGCTCAA------------------GCG-CACCCCC-ACACG--
TCTATTTGACGCTATTGT--
>Peltoboykinia tellimoides
--TCGAAA-CCTGC-C---
TAGCAGAACAACTTTGTGAACATGTAGTT-----ACGCTTGGG-GGGAGAGGA-GTGAAAGCTCT-TTGTCTTCCCAGTGTCGGGA-
TGTGCCCGGTATCTTGTCTC-CATCCAGCGC--TTCACGT--TCGGG-TGAGG--CAATTGCTGGGCA--CC-C---TTGACTT-AACAAC--
AACCCCGGCGTGAATTGCGCCAAGGAA--TTTTATAAA--------AGAGCA--TTCCTCTGTTGCGGTGTA------ATTGCTGTCAA-
TTGGAGGAATTGT--TATCTTTATGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTTTTTGAACGCAAGTTGCGCCMGAAGCCATTTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACACACATCTCGCCA
C-CAACCATTCCCCC--TTGTGGGGTTTTTGAATTTTGTGGCGAGTTGACAT-------TGGTCTCCCG-------TG--
CAAACATTGTTGCGGTTGGCCTAAAATAGAGTACCGA-GTGATGAGATATCACGATAAGTGGTGGTTTAAAAGCCTATTATATGG-
CTTCGCC----AAATCGAGTCGTGAG-TGTCTGTCAC-TTGATGT--ATGCTCAA------------------GAA-AACCCCCCACACA--
TCTAATTGATGCTATTGTCGCGACCCCAGGTCAGGCGGGATT---
>Polygonum hydropiper

---AGGATCATTGTCGAAA-CCTGCAC---
AAGCAG-AAAGACCCGTGAACTCGTTTAC---AAACACCGGGG-G-GATGGCG---------------------TTTGGCCACAAACCAACGACGTCCC------
--CCGAGCTCGG-CAGGAGGC--------CGGCTCCTAGCGAGTCGGTCTCCT-C---CCAGCAC---GAAC-
AAACCCCGGCGCGGATTGCGCCAAGGAC----CATGAACAAT-----AGCGCG-----TCCCACGCCCCTCGGTT-------
GCCCGAAGTGCGCGTGGGCCGTCGTGTCGTTTCGATACATTAGAACGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAA
CGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAAT-
CCGTGAACCATCGAGTCTTTGAACGCAAGTTGCGCCCGAAGCCTTCGGGCCAAGGGCACGTCTGCCTGGGCGTCACGCAC-CGCGTCG-
CCCCC---ACCCCAAACCA---TTGGGATTGGGGGCGGATTG--------------------TGGCCCCCCG-------TG-
CGCTCCTCGCTCGCGGTCGGCCTAAACACAGACCCCGTGGCCGCGAAACGCTGCGACGTTTGGTGGTTTA--------
CTCGTGGCCTTGTGCCTCGAGCATCGCGTCGTGGCCCTGGTGGACC-ATGGGAG-----
CTCAAAGGACCCTGAGGAGGACCGTGTCACCCTCGAGTGGCGTGGGAACCTCCTAACCGTTGCGACCCCAGGTCAGGCGGACTA--
CCGCTGAGTTTAA---
>Rodgersia aesculifolia
--TCGAAA-CCTGC-A---CAGCAG-
AACAACTAGTGAACATGTAGTT-----ACACCTGGG---GAGAGGA-GTTCACGCTCT-TTCTTCC--CGCTGTCAAGA-
TGTGCTCGGTAACTTGTCAC-CCCAAATTGC--TTGACAT---TTTG-GGAAG--GCATTATTGGGCT--CT-C---TTGACTT-AACAAC-
GAACCCCGGCGTGAATTGCGCCAAGGAA--TTT---AAAGAA-----AGAGCA--CTCCTCCATTGTTTTGTGTTCGT----GCAGACAA-
CTGGAGGAAGTGT--TGTATTCTTGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTTTTTGAACGCAAGTTGCGCCCGAAGCCATTTGGTCGAGGGCACGTCTGCCTGGGCGTCACGTACACACATCT-
CCCAC-AAACCTTTCCCCA---TTTTGG-GGGAGGAGT-TTGTGTTGGGAAGACAC-------TGGCCTCCCG-------TG-
CACAACATTGGTGCGGTTGACCTAAAAAAGAGTACCGA-GTGGCGAAATGTCACGATAAGTGGTGGT-TAGAATCC--TAATGTGG--
TTTGCC----AAATCGAGTCGTGAA-CTTTTGTCAC-TCGAGAT--AAGCTCAA-----------------GTG-AACCCCC-ACACG--
TCTAATCGACGCTATG-T--
>Rodgersia podophylla
--TCGAAA-CCTGC-A---CAGCAG-
AACAACTTGTGAACATGTAGTT-----ACACTTGGG---GAGAGGA-GTTCACGCTCT-TTCTTCC--CGCTGTCAAGA-
TGTGCTCGGTAACTTGTCAC-CCCAAATTGC--TTGACGT---TTTG-GGGAG--GCATTATTGGGCT--CT-C---TTGACTT-AACAAC-
GAACCCCGGCGTGAATTGCGCCAAGGAA--TTT---AAAGAA-----AGAGCA--CTCCTCCATTGTTTTGTGTTTGT----GCAGRCAA-
TTGGAGGAAGTGT--TGTCTTCTTGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTTTTTGAACGCAAGTTGCGCCCGAAGCCATTTGGTCGAGGGCACGTCTGCCTGGGCGTCACGTACACACATCT-
CCCAC-AAACCTTTCCCCA---TTTTGG-GGGAGGAGT-TTGTGTCGGGAAGACAC-------TGGCCTCCCG-------TG-
CACAACATTGGTGCGGTTGACCTAAAAAAGAGTACCGA-GTGACGAAATGTCACGATAAGTGGTGGT-TATAATCC--TGATGTGG--
TTTGCC----AAATCGAGTCGTGAA-CGTTTGTCAC-TCGAGAT--AAGCTCAA-----------------GTG-AACCCCC-ACACG--
TCTAATCGACGCTATTGTCGCGACCCCAGGTCAGGCGGGATT---
>Rodgersia sambucifolia

--TCGAAA-CCTGC-A---CAGCAG-
AACAACTTGTGAACATGTAGTT-----ACACCTGGG---GAGAGGA-GTTCACGCTCT-TTCTTCC--CGCTGTCAAGA-
TGTGCTCGGTAACTTGTCAC-CCCAAATTGC--TTGACAT---TTTG-GGAAG--GCATTATTGGGCT--CT-C---TTGACTT-AACAAC-
GAACCCCGGCGTGAATTGCGCCAAGGAA--TTT---AAAGAA-----AGAGCA--CTCCTCCATTGTTTGTGTTCGT----GCAGACAA-
CTGGAGGAAGTGT--TGTCTTCTTGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTTTTTGAACGCAAGTTGCGCCCGAAGCCATTTGGTCAAGGGCACGTCTGCCTGGGCGTCACGTACACACATCT-
CCCAC-AAACCTTTCCCCA---TTTTGG-GGGAGGAGT-TTGTGTTGGGAAGACAC-------TGGCCTCCCG-------TG-
CACAACATTGGTGCGGTTGACCTAAAAAAGAGTACCGA-GTGGCGAAATGTCACGATAAGTGGTGGT-TAGAATCC--TAATGTGG--
TTTGCC----AAATCGAGTCGTGAA-CGTTTGTCAC-TCGAGAT--AAGCTCAA------------------GTG-AACCCCC-ACACG--
TCTAATCGACGCTATTGTCGCGACCCCA---
>Saxifraga atrata
--ATGAAA-CCTGCCA---AAGCAG-
AAAACATCGAGAACATGTAAGA---AAAAAATTGAG-G-GGGAGCA-TTCTATGCTTC-TTCTCAC--CTTTGTCGGGA-
GGCATTAGATTCATTACTCT-CTGCG-TTGC-TCTTACGC--ACGTG-GGAGT--AAGT-ATCGAGTG--TTTC---CTTACAT-
TGCAACAAACCCCCGGCGTGAATTGCGCCAAGGAA---TTTAAAAAAAAGAAAGATAGCA--TTCCTCCATCT---CATGT------------------
TGGAGAAAATGT--TATCTTCTTGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTTTTTGAACGCAAGTTGCGCCCAAAGCCACCTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACACACTTCT-
CCCAC-AAACCTTTCCCAA---TTATGG-TAAGGGGAA-TTGTGGTGAGTAGAGAT-------TGGTCTCCCA-------TG-CCC---
TTGCGTGTGGTTGGCCTAAAAAAGAGTA-CGA-GTGATGAAACGTCACGATTAGTGGTGGTATATAAGCC--TCTTTTGG-TTTTGCC----
AGATCGAGTCGAA---CGTTTGTCAC-TTGAGAT--ATGCTCTA------------------ATT-AATCCCC-GAACG--TTGAATTGACGCTATT-T------------
----
>Saxifraga clusii
---TGCTGAAA-CCTGC-A---ATGCAG-
AAAACCGTGTGAACGCGTGACC---TTAAAAAAGAG-A-AGGGGCA-TTCTTTGTTCC-TTTTTAC--CTTTGTTGGAT--TTGCTTCATTCCTTT-
GTC-GTGTGCTTGC-TTCCATGC--ATGCA-CGTCG--GAGT-ATTGAGTG--TT-C---CCTACAA-TGTAAC-
AACCCCCGGCGCGAATTGCGCCAAGGAATTTTTAAAAAAAG-----AGGGCA-CTTTCTCCACTT---CATGCC----AT-GCACAAAG--
TGGAGAGAATGT--TATCTTCTAGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTTTTTGAACGCAAGTTGCGCCTGAAGCCTCTTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACACACTTCT-CCCAC-
AATCC-TTTCACA----TGTAG-TGTTTGCGT-TTGCGGCGAGGGATTAT-------TGGCCTCCCA-------TG-CCC---
ATGTGTGTGGTTGGCCTAAAAATGAGTA-CTA-GTGATGAAACGTCACGATAAGTGGTGGTACATAAGCC--TTCTTTGG-TTTTGCC----
AAATCGAGTCGTA---AGTTTGTTGC-TTGAGAA--ATACTCAR------------------GTT-AATCCCC-TTACG--TTGATCATACGCTATT-
TCGCGACCCCAGGTCAGGCGGGATTACCCGCTGAGTTTAAGCATATCAATAA---
>Saxifraga fortunei

---CTGAAA-CCTGC-AATGATGCAG-
AAAACCTTGAGAACATGTAAAA---AAAATATTGAG-G-AGGGGCA-TTTCATGTTTC-TTCTCAC--CTTTGTTGGGT-
GTCATTCAATTTTTTTAACC-TTGCATTTGC-TTTCACGCATGTATG-GGTGT--AAATAATTGAGTG--TT-C---CTTACAT-TGCAAC-
AACCCCCGGCGTGAATTGCGCCAAGGAA-TTTTTAAAAAAAG-----AGAGCA--TTTCTCCATTT---CATGTGTAT----GCATTATT--
TGGAAAAAAATGT--TATCTTATTTATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTTTTTGAACGCAAGTTGCGCCCGAAGCC-TCTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACACACTTCT-CCCAC-
AAACC-TTTCCCA---TTTTGG-TTTAGGCAT-TTGTGATGAGAAAAGAT-------TGGCCTCCCA-------TA-
CCCTCCTTGCGTGTGGTTGACCTAAAAAAGAGTA-CGA-GTGATGAAACGTCACGATAAGTGGTGGTTTATAAGCC--TTTTTTGG-
TTTTGCC----AAATCGAGTCGAA---CGTTTGTCAC-TTGAAAT--ATGCTCAA------------------GTT-AATCCCC-ACACG--TG----------------------------
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>Saxifraga melanocentra
---ATGCAA-CCTGCCA---AAGCAG-
AAAACATCGAGAACATGTAACA-----AAAATTGAG-T-GGGAGCA-TTCCATGCTTC-TTCTCAC--
CTTTGTCGGGAGGGCATTAGATTCATTGCTCC--TGCG-TTGC-TCTTACGC--ACGTG-GGAGT--AAGT-ATCGAGTG--TTTC---CTTACAT-
TGCAACAAACCCCCGGCGTGAATTGCGCCAAGGAATTTTA---AAAAAAGATAGATAGCA----CCTCCATCT---CATGTT------------------
GGAGAAATGT--TATCTTCTTGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTTTTTGAACGCAAGTTGCGCCCGAAGCCACCTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACACACTTCT-
CCCAC-AAACCTTTTCCAA---TTATGG--TGAGGCAT-TTGTGGTGAGTAGAGAT-------TGGTCTCCCA-------TG-CCC---
TTGCGCGTGGTTGGCCTAAAAAAGAGTA-CGA-GTGATGAAACGTCACGATTAGTGGTGGTATATAAACC--TCTTTTGG-TTTTGCC----
AGATCGAGTCGAA---CGTTTGTCAC-TTGAGAT--ATGCTCTA------------------GTT-AATCCCC-GAACG--TTGAATTGACGCTATTTT-------------
----
>Saxifraga pallida
---ATGAAA-CCTGCCA---AAGCAG-
AAAACATCGAGAACATGTAACA---AAAAAATTGAG-A-TGGAGCA-TTTCATGCTTC-TTCTCAC--CTTTGTCGGGA-
GGCATTAGATTCATTACTCT-CTGTG-TTGC-TCTTACGC--ACGTG-GGAGT--AAGT-ATCGAGTG--TTTC---CTTACAT-
TGTAACAAACCCCCGGCGTGAATTGCGCCAAGGAA---CTTTAAAAAAAAGATAGATAGCA--TTCCTCCATCT---CATGT------------------
TGGGGAAAATGT--TATCTTCTTGATGTCTT-
TAATGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTGAATTGCAGAATCCCGT
GAACCATCGAGTTTTTGAACGCAAGTTGCGCCTGAAGCCACCTGGTTGAGGGCACGTCTGCCTGGGCGTCACGTACACACTTCT-CCCAC-
-AACCTTTCCCAA---TTATGG-TTAGGGAAA-TTGTGGTGAGTAGAGAT-------TGGTCTCCCA-------TG-CCC---
TTGCGTGTGGTTGGCCTAAAAAAGAGTA-CGG-GTGATGAAACGTCACGATTAGTGGTGGTATATAAGCC--TCTTTTGGTTTTTGCC----
AGATCGAGTCGAA---CGTTTGTCAC-TTGAGAT--ACGCTCTA------------------GTT-AATCCCC-GAACG--TTGAATTGACGCTATTTT------------
----
>Saxifraga stellaris

---CTGAAA-CCTGC-A---ATGCAG-
AAAACCGTGTGAACACGTGACC---TCAAAAAAGAG-A-AGGAGCA-TTCTTTGTTCCTTTTTTAC--CTTTGTTGGAT--
TCGCTTCATTCCTTT-GTC-GTGTGCTTGC-TTCCATGC--ATGCA-CGTCG--GAGT-ATTGAGTG--TT-C---CCTACAA-TGTAAC-
AACCCCCGGCGCGAATTGCGCCAAGGAA--TTTTTAAAAAAG-----AGAGCA--TTTCTCCACTT---CATGCC----AT-GCACAAAG--
TGGAGAGAATGT--TATCTTCTAGAT--

>Urtica dioica
---TTTCCGTAGGTGAACCTGCGGAAGGATCATTGTCGAA--
CCTGC--TTCATGCAA-AATGACCCGCGAATAAGTTCTT---ACGTTTTGGGGCA-AGGATGG----------------------CT----------
CGTACCCGACCATTCTTGTC-------TTGC--TT-C------------TAAC-AACCAAAGGCGCGGGATGCGCCAAGGAA-
-AATCCAAACGAG-----TTTGACTCTTGCCTCGGTGCATTGCATCGTG----GCAGCGAG----------------TGTATTCGATAAGTCGT--
AACGACTCTCGGCAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCAAAATGCGATACGTGGTGTGAATTGCAGGATCCCGTG
AACCATCGAGTTTTTGAACGCAAGTTGCGCCCGAAGCCTTTAGGCCGAGGGCACGTCTGCCTGGGCGTCACG----
CACCGTTGCCCTCCAAACC--TCCGCAGTCCTTTAG---
TGGGATTGTTGAGATGTGCGGGGGTCGTAAAGTGGCTTCCCGTCGGCTTTGTCCC---------GCGGTTGGCCTAAAAATGTATCCCTA-
GTCGCGGTGCG-CGCGGCATTCGGTGGTC----------ATCGATAGATTTCGTT------ACCCCGCCGTG---CGTTC-
CCGTGTTGCGAAGGATGTTAGC------------------AGC-AAACCC---GATG--TCT------CGCT----CTGTG---
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APPENDIX B

MOLECULAR ACCESSIONS USED FOR THE PRESENT STUDY (GENBANK)

1 gi|532159323|gb|KF196317 1| Micranthes ferruginea isolate SaxITS1 18S ribosomal RNA gene, partial sequence; internal transcribed spacer 1, 5 8S ribosomal RNA gene, and internal transcribed spacer 2, complete sequence; and 28S ribosomal RNA gene, partial sequence
2 gi|809277889|emb|LM654392 1| Micranthes petiolaris genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher D Gvarts 270 (NY)
3 gi|809277843|emb|LM654346 1| Micranthes bryophora genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher P L Packard & N Tobias 78-281 (NY)
4 gi|809277842|emb|LM654345 1| Micranthes bryophora genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher B Ertter 6737 (NY)
5 gi|809277855|emb|LM654358 1| Micranthes foliolosa genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher V V Petrovsky, T M Koroleva (LE)
6 gi|809277854|emb|LM654357 1| Micranthes ferruginea genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher A G Jones 5577 (LE)
7 gi|809277896|emb|LM654399 1| Micranthes redofskyi genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher A I Tolmachev, T G Polozova (LE)
8 gi|809277863|emb|LM654366 1| Micranthes laciniata genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher Tson The Son (LZ3762)
9 gi|809277903|emb|LM654406 1| Micranthes stellaris genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher M Roeser 6226 (HAL)
10 gi|495021299|gb|KC749987 1| Micranthes stellaris voucher FCO:32853 internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence
11 gi|809277904|emb|LM654407 1| Micranthes stellaris genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher N Tkach
12 gi|495021297|gb|KC749985 1| Saxifraga clusii voucher FCO:32851 internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence
13 gi|495021298|gb|KC749986 1| Saxifraga clusii subsp lepismigena voucher FCO:32856 internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence
14 gi|809277884|emb|LM654387 1| Micranthes occidentalis genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher P Lesica 3712 (NY)
15 gi|809277853|emb|LM654356 1| Micranthes fallax genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher s coll (L-W1997BL00485)
16 gi|809277861|emb|LM654364 1| Micranthes howellii genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher R R Halse 7427 (NY)

17 gi|809277852|emb|LM654355 1| Micranthes eriophora genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher Lehto, Hensel, Pinkava 10853A (NY)
18 gi|809277888|emb|LM654391 1| Micranthes pensylvanica genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher N Tkach & M Roeser (A/0285)
19 gi|809277911|emb|LM654414 1| Micranthes virginiensis genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher R Kral 94766 (NY)
20 gi|809277893|emb|LM654396 1| Micranthes purpurascens genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher L Malyshev (LE)
21 gi|809277900|emb|LM654403 1| Micranthes rufidula genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher C L Hitchcock & C V Muhlick 21494 (NY)
22 gi|809277872|emb|LM654375 1| Micranthes micranthidifolia genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher A Bond 64 (NY)
23 gi|809277867|emb|LM654370 1| Micranthes marshallii genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher R R Halse 4003 (NY)
24 gi|809277844|emb|LM654347 1| Micranthes californica genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher P Elvander 834 (NY)
25 gi|573005990|gb|KC691723 1| Micranthes foliolosa 5 8S ribosomal RNA gene, partial sequence; internal transcribed spacer 2, complete sequence; and 28S ribosomal RNA gene, partial sequence
26 gi|809277899|emb|LM654402 1| Micranthes rhomboidea genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher K H Lackschewitz 4238 (NY)
27 gi|809277862|emb|LM654365 1| Micranthes integrifolia genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher T Tanaka & C Richardson 49 (NY)
28 gi|809277886|emb|LM654389 1| Micranthes oregana genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher B Lesica 8686, C Bjork (NY)
29 gi|809277876|emb|LM654379 1| Micranthes nelsoniana genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher M H Hoffmann 11/39 (HAL)
30 gi|809277892|emb|LM654395 1| Micranthes purpurascens genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher Antonova (LE)
31 gi|809277885|emb|LM654388 1| Micranthes odontoloma genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher E Neese 15194 (LE)
32 gi|809277873|emb|LM654376 1| Micranthes nelsoniana genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher B A Jurtzev, V V Petrovsky (LE)
33 gi|809277845|emb|LM654348 1| Micranthes calycina genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher E V Zimarskaya, A A Korobkov, B A Jurtzev (LE)
34 gi|809277846|emb|LM654349 1| Micranthes calycina genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher S Kharkevich & V Barkalov 429 (NY)

35 gi|809277898|emb|LM654401 1| Micranthes reflexa genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher M H Hoffmann 11/23 (HAL)

36 gi|809277895|emb|LM654398 1| Micranthes razshivinii genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher C B Hardham 21024 (NY)

37 gi|809277894|emb|LM654397 1| Micranthes razshivinii genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher M H Hoffmann 11/36 (HAL)

38 gi|809277877|emb|LM654380 1| Micranthes nelsoniana subsp aestivalis genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher D Shaulo & E Shulgina 66 (LE)

39 gi|809277839|emb|LM654342 1| Micranthes aprica genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher M F Denton 4238 (NY)

40 gi|809277897|emb|LM654400 1| Micranthes reflexa genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher S L Welsh & J K Rigby 11302 (LE)

41 gi|809277878|emb|LM654381 1| Micranthes nelsoniana var pacifica genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher A V Smirnov (LE)

42 gi|809277906|emb|LM654409 1| Micranthes tempestiva genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher K L Lackschewitz 3806 (NY)

43 gi|809277901|emb|LM654404 1| Micranthes sachalinensis genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher E Pobedimova & G Konovalova 680 (LE)

44 gi|809277890|emb|LM654393 1| Micranthes porsildiana genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher V I Dorofeev 154 (LE)

45 gi|809277879|emb|LM654382 1| Micranthes nidifica genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher K H Lackschewitz 4819 (NY)

46 gi|809277875|emb|LM654378 1| Micranthes nelsoniana genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher A A Galanin, L P Gorbunova, S A Shabarshina (LE)

47 gi|809277874|emb|LM654377 1| Micranthes nelsoniana genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher N V Matveeva & l L Zanokha 3463 (LE)

48 gi|809277881|emb|LM654384 1| Micranthes nivalis genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher L B Jorgensen & S Larsson 66-2437 (NY)

49 gi|809277902|emb|LM654405 1| Micranthes spicata genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher E Hulten (NY)

50 gi|809277909|emb|LM654412 1| Micranthes tilingiana genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher S Kharkevich, A Bobrov (JE)

51 gi|809277907|emb|LM654410 1| Micranthes tenuis genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher N V Matveeva & l L Zanokha 3423 (LE)

52 gi|809277848|emb|LM654351 1| Micranthes caroliniana genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher S A Spongberg, D E Boufford 17107 (LE)

53 gi|809277847|emb|LM654350 1| Micranthes careyana genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher S A Spongberg & D E Boufford 17104 (LE)

54 gi|809277838|emb|LM654341 1| Micranthes apetala genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher C L Hitchcock & C V Muhlick 22427 (NY)
55 gi|809277866|emb|LM654369 1| Micranthes lyallii subsp hultenii genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher J A Calder 32273, D B O Savile (LE)
56 gi|809277865|emb|LM654368 1| Micranthes lyallii genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher J A Calder 11236, D B O Savile (LE)
57 gi|809277908|emb|LM654411 1| Micranthes texana genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher R L McGregor 17456 (NY)
58 gi|363548361|gb|JN375576 1| Saxifraga fortunei var alpina 18S ribosomal RNA gene, partial sequence; internal transcribed spacer 1, 5 8S ribosomal RNA gene, and internal transcribed spacer 2, complete sequence; and 28S ribosomal RNA gene, partial sequence
59 gi|809277869|emb|LM654372 1| Micranthes melanocentra genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher G & S Miehe 00-246-10 (Marburg, Germany)
60 gi|809277887|emb|LM654390 1| Micranthes pallida genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher G & S Miehe 00-205-22 (Marburg, Germany)
61 gi|809277857|emb|LM654360 1| Micranthes fusca genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher S S Ikonnikov 27 (LE)
62 gi|809277856|emb|LM654359 1| Micranthes fusca genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher N A Brummit & G Firsov 180 (LE)
63 gi|809277840|emb|LM654343 1| Micranthes atrata genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher Guo Benzhao 9318 (KUN0255501)
64 gi|809277851|emb|LM654354 1| Micranthes divaricata genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher A Favre & S Matuszak 190 (LZ)
65 gi|809277905|emb|LM654408 1| Micranthes subapetala genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher K L Lackschewitz 5346 (NY)
66 gi|809277859|emb|LM654362 1| Micranthes hieraciifolia genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher N Tkach
67 gi|809277860|emb|LM654363 1| Micranthes hieraciifolia subsp longifolia genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher A A Korobkov, N A Sekretareva (LE)
68 gi|809277864|emb|LM654367 1| Micranthes lumpuensis genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher G & S Miehe & U Wuendisch 94-433-18 (Marburg, Germany)
69 gi|163433857|gb|EU158863 1| Saxifraga pallida internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence
70 gi|163433840|gb|EU158846 1| Saxifraga melanocentra internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence
71 gi|387935837|gb|JN102225 1| Saxifraga pallida voucher Ying et al 1342 internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence
72 gi|163433846|gb|EU158852 1| Saxifraga atrata internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence

73 gi|169635893|dbj|AB248848 1| Rodgersia podophylla genes for ITS1, 5 8S rRNA, ITS2, 26S rRNA, complete and partial sequence, strain: ROD01
74 gi|159795892|gb|EU239674 1| Bergenia purpurascens isolate R10 18S ribosomal RNA gene, partial sequence; internal transcribed spacer 1, 5 8S ribosomal RNA gene, and internal transcribed spacer 2, complete sequence; and 28S ribosomal RNA gene, partial sequence
75 gi|573005989|gb|KC691722 1| Micranthes hieraciifolia 5 8S ribosomal RNA gene, partial sequence; internal transcribed spacer 2, complete sequence; and 28S ribosomal RNA gene, partial sequence
76 gi|444289530|gb|JQ895206 1| Rodgersia podophylla voucher Sun 0704, Nikko Botanical Garden internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence
77 gi|444289545|gb|JQ895221 1| Astilboides tabularis voucher Zhu 040 (KUN) internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence
78 gi|14210115|gb|AF374827 1| Saxifraga stellaris internal transcribed spacer 1, partial sequence
79 gi|444289529|gb|JQ895205 1| Rodgersia sambucifolia voucher Zhu 041 (KUN) internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence
80 gi|444289544|gb|JQ895220 1| Bergenia purpurascens voucher Zhu 0601 (KUN) internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence
81 gi|387935844|gb|JN102232 1| Rodgersia aesculifolia voucher Ying et al 1668 internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence
82 gi|387935842|gb|JN102230 1| Bergenia purpurascens voucher Lei KBG01 internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence
83 gi|809277868|emb|LM654371 1| Micranthes melaleuca genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher A Maneev, A Krasnikov, I Krasnoborov (LE)
84 gi|387935843|gb|JN102231 1| Oresitrophe rupifraga voucher Lei KBG02 internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence
85 gi|169635957|dbj|AB292041 1| Darmera peltata genes for ITS1, 5 8S rRNA, ITS2, complete sequence
86 gi|169635892|dbj|AB248847 1| Peltoboykinia tellimoides genes for ITS1, 5 8S rRNA, ITS2, 26S rRNA, complete and partial sequence, strain: PEL01
87 gi|809277841|emb|LM654344 1| Micranthes brachypetala genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher V Siplivinsky (LE)
88 gi|809277891|emb|LM654394 1| Micranthes punctata genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher E V Dorogostayskaya (LE)
89 gi|747154876|gb|KM887391 1| Bergenia ligulata isolate SBB-1380 internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence
90 gi|747154871|gb|KM887386 1| Bergenia ligulata isolate SBB-1379 internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence
91 gi|809277850|emb|LM654353 1| Micranthes davurica var NT-2014 genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher M P Andreev, E Yu Norkina, V V Petrovsky (LE)
92 gi|809277882|emb|LM654385 1| Micranthes nudicaulis genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher B A Jutzev (LE)

93 gi|338857204|gb|JF980321 1| Chrysosplenium japonicum isolate P307 18S ribosomal RNA gene, partial sequence; internal transcribed spacer 1, 5 8S ribosomal RNA gene, and internal transcribed spacer 2, complete sequence; and 26S ribosomal RNA gene, partial sequence

94 gi|444289570|gb|JQ895246 1| Peltoboykinia tellimoides voucher Sun 0701, Nikko Botanical Garden internal transcribed spacer 1, partial sequence; 5 8S ribosomal RNA gene, complete sequence; and internal transcribed spacer 2, partial sequence

95 gi|809277883|emb|LM654386 1| Micranthes oblongifolia genomic DNA containing ITS1, 5 8S rRNA gene, ITS2, specimen voucher A A Pirozhenko (LE)

96 gi|169635894|dbj|AB248849 1| Elmera racemosa genes for ITS1, 5 8S rRNA, ITS2, complete sequence, strain: EL02

97 gi|54289839|dbj|AB163497 1| Mitella japonica genes for ITS1, 5 8S rRNA, ITS2, complete sequence, clone:GIVb gi|54289840|dbj|AB163498 1| Mitella japonica genes for ITS1, 5 8S rRNA, ITS2, complete sequence, clone:GIVc

98 gi|54289838|dbj|AB163496 1| Mitella japonica genes for ITS1, 5 8S rRNA, ITS2, complete sequence, clone:GIVa

99 gi|363548340|gb|JN375555 1| Chrysosplenium sinicum 18S ribosomal RNA gene, partial sequence; internal transcribed spacer 1, 5 8S ribosomal RNA gene, and internal transcribed spacer 2, complete sequence; and 28S ribosomal RNA gene, partial sequence

100 gi|363548339|gb|JN375554 1| Chrysosplenium sinicum 18S ribosomal RNA gene, partial sequence; internal transcribed spacer 1, 5 8S ribosomal RNA gene, and internal transcribed spacer 2, complete sequence; and 28S ribosomal RNA gene, partial sequence